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**THE DISTRIBUTION AND ACTIVITIES  
OF BACTERIA IN SOILS OF  
THE ARID REGION**

**BY**

**CHARLES B. LIPMAN**

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# THE DISTRIBUTION AND ACTIVITIES OF BACTERIA IN SOILS OF THE ARID REGIONS\*

BY

CHARLES B. LIPMAN

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## INTRODUCTION

The student of soils in the humid region, when for the first time exploring soils in the arid region, is invariably struck with the extraordinary depth of the latter as against the very shallow nature of the former. Taken by and large, and excepting the faulty soils, including those underlaid at no great depth by stiff clay, coarse gravel, hardpan, or original rock, respectively, the soils of the arid region very commonly show a depth of at least eight to ten feet, and, when viewed in section, exhibit such a striking uniformity in texture and color as to attach to this unusual condition, in the mind of the observer, a certain marked practical and scientific interest. The full significance to crops of the arid region of this extraordinary condition in our soils was first realized and pointed out by Hilgard and was made the subject, by him and Loughridge, of a comprehensive investigation on the "soil-columns" of California, a large part of which is completed, but some of which is still in progress. The study of the soil-columns of California comprised what might be looked upon as a very thorough partial soil survey of California. It was the intention of the investigators above named, at the inception of the work, to obtain columns of soil representing depths of twelve feet, including a sample for every foot

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\* Read before the Society of American Bacteriologists, Washington, D. C., December 27, 1911.

in depth, and to obtain a knowledge of the chemical constitution and the texture of the soils by making systematic chemical and mechanical analyses of all the samples thus collected. The information thus obtained in the several years in which the soil-columns were studied by Hilgard and Loughridge and the large number of types of soils considered, along with the most striking circumstance of the depths to which plant root-systems of the arid regions penetrated, led Hilgard to believe that the striking chemical and mechanical differences between the soils of the arid and humid regions, as well as the differences in the development of the root-systems in these regions, respectively, might find a parallel also in a difference between the bacterial flora at various depths in the soil. It was this belief on Hilgard's part and his valuation thereof as being of exceeding scientific interest as well as practical value, that led to the association with him something over three years ago of the writer, and it was then that I undertook, among other biological problems in soils, a study of the nitrogen-transforming and nitrogen-fixing bacteria in the different layers of soils in the arid region. This study, while it has progressed considerably, is still in the first stage of its development and the complete results thereof are intended ultimately to be combined with the mechanical and chemical analyses of these soils in a comprehensive report on the whole work. For the purposes of this paper, it is sufficient to give a resumé of some of the important results obtained in these investigations with an account of the methods employed in the work, so that it may serve as a preliminary communication on the subject and bring out certain striking facts with reference to the distribution of bacteria in California soils.

## METHODS EMPLOYED IN THESE INVESTIGATIONS

One of the most difficult problems in connection with these investigations was to find a method for the collection of soil samples at the several depths which would fairly represent the actual conditions which obtain there, so far as the bacterial flora are concerned. Our first attempts in this direction were made with an auger of the type manufactured by Iwan Brothers at South Bend, Indiana, by means of which we tried, through

successive sterilization of the auger (before taking each sample), to obtain a sample which represented, uncontaminated, each of the soils as they are found in their natural state in the field. With a sterile spatula there were taken from the samples thus obtained with the ~~auger~~ representative samples which were immediately placed in sterile cotton-stoppered bottles. It was soon found, however, that this method could not be relied upon for accurate results, since no matter how carefully the samples were thus taken, there were many chances for contaminating samples from the lower layers with soil from the upper layers and thus obtaining results which were erroneous. After much experimenting we finally decided on the following plan for taking the soil samples, which, so far as I know, is as free from chances of error as any method that can be adopted in a series of investigations which must of necessity be so extensive. Indeed, I believe the chances for error here are so small that they cannot affect the validity, to any appreciable extent, of the results obtained. Our method consists in having dug, a day or two prior to sampling, a hole twelve feet in depth with at least one vertical wall and large enough for a man to stand in. The samples are taken as follows: With a sterilized spade, a layer of soil of about five or six inches in depth is sliced down along the whole length of the wall which is to be sampled. After this is done, to remove the soil that in the one or two days' exposure may have become contaminated, the fresh surface thus obtained on the vertical wall is sterilized by means of a plumber's torch on the surface surrounding every spot, previously marked off, at which a sample from each foot in depth is to be taken. When this is done a sterile cylindrical tin tube, a little over one inch in diameter and about ten inches long, is driven at right angles to the wall into the spot selected for sampling, immediately drawn out when sufficient soil has thus been obtained, and the cotton plug replaced. In our first experiments, glass tubes of the size described were employed, with paraffined corks at one end and cotton stoppers at the other. We found this to be a poor method, however, and have replaced the glass by tin tubes, closed at one end and plugged with cotton at the other. These are sterilized at 150 degrees centigrade for one hour and a half



before using. In this way by the use of a plumber's torch at every depth as we descend from the surface of the soil down to the twelve-foot depth, we obtain, by starting at sterile surfaces, a sample of soil representing as nearly as possible the true condition which obtains at very depth. The samples are marked properly, taken to the laboratory, and examined for their ammonifying, nitrifying and nitrogen-fixing powers by means of a modified Remy method, the solutions employed for the work being prepared in accordance with the formulae used by J. G. Lipman.<sup>1</sup> Every 50 c.c. portion of the medium in a 250 c.c. Erlenmeyer flask is inoculated with 5 grams of soil.

## DESCRIPTION OF SOILS EMPLOYED IN THESE EXPERIMENTS

The descriptions given below represent the soils which were employed for bacteriological examinations and sampled for the purpose as above described. The numbers employed below are used throughout all the following tables so as to make unnecessary any further descriptions.

Soil No. 1. Red clay loam mesa soil, from Riverside, California, on which good orange trees were growing at time of sampling. The soil is well supplied with potash, but rather poor in phosphoric acid and very poor in humus and nitrogen. It is underlaid by hardpan at six feet from the surface, which continues on down to the twelve-foot depth. With the careful cultivation which is given it, along with proper fertilization and tillage, the soil produces profitable crops of oranges and lemons.

Soil No. 2. Silty alluvial loam, from Davis, California. The samples used were obtained from between some fig trees at the University Farm. This soil is practically uniform in color from the first foot to the twelfth and only becomes slightly different in texture below the fifth foot, becoming gradually coarser and sandier as we descend to the lower layers. It is well supplied with potash, phosphoric acid, and lime and has, for a soil of the arid region, a normal content of humus.

Soil No. 3. Sandy alluvial loam, from Davis, California. Samples were taken from a wheat field at the University Farm, only to a depth of ten feet. This soil is well supplied with phosphoric acid, potash and lime, but rather poor in humus and nitrogen. The sand is of a coarse nature and becomes rapidly coarser, descending from the first foot down to the twelfth, where it is found as very coarse sand.

<sup>1</sup> Bulletin 180. N. J. Agr. Expt. Station.

Soil No. 4. Sandy alluvial loam, from Davis, California. Samples obtained in almond orchard at the University Farm. This soil is not nearly so coarse as soil No. 3 and shows a more uniform texture throughout a seven-foot depth, but after that becomes coarser in texture. It is better supplied with humus and nitrogen than soil No. 3 and is well supplied with potash, phosphoric acid, and lime.

Soil No. 5. Alluvial loam, from Davis, California. Samples obtained in a pear orchard at the University Farm. The soil is uniformly of a fine sandy loam texture for a depth of nearly five feet and then rapidly becomes much coarser than the soil at similar depths in No. 3. The upper soil is well supplied with potash, phosphoric acid, and lime, and fairly well supplied with humus and nitrogen. The lower layers are rather poor in phosphoric acid, humus and nitrogen.

Soil No. 6. Fine silty soil, from Hanford, California. Samples taken from a vineyard at Hanford, from the first to the ninth foot only. No sampling was done below the ninth foot because of the fact that the water-table was reached at about that point and it was almost impossible to get samples uncontaminated. This soil is almost devoid of humus and contains but little nitrogen, but is fairly well supplied with phosphoric acid, potash, and lime. No alkali is present in the soil.

Soil No. 7. Silty alluvial loam, from Davis, California. Samples taken at the University Farm, close to a young eucalyptus tree, about twenty months old. The soil is fairly uniform in texture throughout the entire depth studied and is fairly well supplied with humus and nitrogen, and well supplied with phosphoric acid, potash, and lime. No alkali is present.

Soil No. 8. Alkali soil from Tulare, California. Taken only to a depth of ten feet, owing to water conditions such as those described in soil No. 6. This soil contains very little humus and is strongly impregnated with salts, especially "black alkali." It is otherwise well supplied with phosphoric acid, potash, lime, and the other minerals. Hardly any vegetation can exist on this soil after the salts have risen to the upper layers.

Soil No. 9. A very stiff and tenacious silty clay adobe, from Imperial, California. Uniform in texture from the surface down to the eighth foot, at which there is found a layer of fine sand for a foot and a half in depth and then a silty sand below to the twelve-foot depth. The soil throughout is almost devoid of humus and contains but very little nitrogen. It is very rich, however, in phosphoric acid, potash, and lime. The upper layers of the soil consist of particles of silt and clay which are so fine as to become cemented together into an extremely hard, refractory material, which is almost of the consistency of a dry, but not heated, brick. A considerable quantity of common salt is present in this soil. This soil has never been cultivated or cropped.

Soil No. 10. Fine, sandy soil from the desert of Coachella Valley. In this, as in the Imperial Valley soils, there are to be found narrow layers of an inch or two, and sometimes more, of very fine shells of former life which existed in the water at one time covering this land. The soil has very

little or no humus and nitrogen. It is, however, rich in phosphoric acid and lime and well supplied with potash. The soil is uniform in texture throughout the twelve-foot depth and becomes only a little coarser at twelve feet. The only changes visible in color and texture in the vertical wall are merely those of the shell layers above noted. The soil from which these samples were taken has never been cropped, but similar soil, with a good water supply, produces very fine alfalfa. Very little alkali is present in this soil.

Soil No. 11. Fertile, alluvial loam from Hayward, California. Uniform in texture for seven feet and then rapidly becoming quite coarse and remaining so down to a depth of twelve feet. This soil is very fertile, producing good crops of cherries, walnuts, potatoes, and other agricultural plants. It is well supplied with humus and nitrogen, judged by the standard for soils of the arid region, throughout the twelve-foot depth. The phosphoric acid, potash, and lime are also plentiful in all the soil layers. The samples used in these investigations were obtained in a cherry orchard.

## AMMONIFICATION IN SOIL COLUMNS

Second only to the importance of soil bacteria in maintaining the total nitrogen supply in soils is their power to supply constantly available nitrogen to plants. The essential nature of this important phase of the activities of soil organisms is in no wise detracted from by the recent research which has made it clear that some plants at least can take their nitrogen from the soil in forms other than the nitrate. While many of them may not absorb their nitrogen in the form of nitrates, it seems quite certain that practically all of them must take their nitrogen in forms much simpler than the proteid. This being undeniably the case, some agency in the soil is necessary to accomplish the transformation of the organic nitrogen (no matter what the source of the latter to the soil may be) into a simpler, more available, or more assimilable form. These agencies we have found to be the various types of soil organisms which constitute what we now designate by the term "ammonifying flora" of the soil.

With these statements admitted, it seems reasonable to suppose that any increase in the activities of the organisms, included under this head, is a distinct advantage to the plant. Under our climatic conditions, where, as above stated, the plant roots very deeply, besides making a large lateral root-development, it is necessary to have the activities of the ammonifying organisms

not only in the upper layers of the soil, but in the lower layers where an actual examination of the root-systems of plants shows a large development of fibrous or feeding roots. A study, therefore, of the ammonifying powers of the different layers of soil, or, rather, of the microorganic flora which they contain, is of practical moment, since it is bound to throw light on the soluble nitrogen supply for roots in the greater depths of soil and indicate what practical measures may be taken toward sustaining and encouraging the growth and activities of the organisms responsible for that soluble nitrogen supply. Since, therefore, we assume ammonia production to be the first great step recognized by our analytical methods in the transformation of soil nitrogen, I have first determined the ammonifying powers at various depths of soils, which may be considered typical of well-defined areas and conditions in the arid region.

For this purpose there were inoculated into sterile 50 c.c. portions of 1 per cent peptone solution, 5 grams of soil from every foot from the surface down to the last depth taken, as above described. After four days incubation at about 28 degrees centigrade, the cultures were washed into copper distilling flasks. sufficient distilled water added, as well as a slight excess of magnesia, and distilled. The distillate was caught in standard tenth normal hydrochloric acid, the excess of which was titrated with standard tenth normal ammonia. Table I gives the results of determinations of the ammonifying power of the soils chosen, as above described. The ammonifying power of only one soil, namely No. 6, is not given, for the reason that the soil column had inadvertently become contaminated before we were ready to use it.

The numbers of the soils refer to corresponding numbers under the descriptions given above, and the amounts of ammonia produced, as given in the table, represent milligrams of nitrogen as ammonia.

The data given in table I prove very clearly two facts. First, that in the typical deep and normal soils of the arid region, the activities and the distribution of the ammonifying flora seem to run parallel with the texture, the chemical composition, and the root-development in these soils. Second, that in the absence of

TABLE I  
AMMONIFICATION IN SOIL COLUMNS

Soil No.	1	2	3	4	5	6	7	8	9	10	11
	mg.	mg.	mg.	mg.	mg.		mg.	mg.	mg.	mg.	mg.
1st ft.	59.80	67.76	68.25	61.60	54.67		72.05	7.28	25.46	12.15	42.84
2nd ft.	55.46	77.00	72.66	54.46	49.14		68.90	4.90	8.27	6.46	36.84
3rd ft.	52.30	69.02	68.40	38.50	32.76		70.43	5.46	8.20	3.21	14.14
4th ft.	55.83	70.63	43.05	44.80	lost		65.72	2.80	8.69	1.99	7.28
5th ft.	49.72	67.48	34.23	39.90	8.40		63.69	4.76	4.87	1.25	7.84
6th ft.	49.00	84.63	31.64	47.60	15.68		60.50	4.41	8.27	1.61	21.19
7th ft.	31.70	72.17	42.00	44.80	17.85		56.90	2.45	6.03	1.57	7.14
8th ft.	30.54	52.57	35.00	22.54	9.80		50.43	10.22	5.40	1.34	7.48
9th ft.	28.65	52.43	35.70	32.90	15.40		45.69	10.64	2.18	1.24	5.60
10th ft.	25.40	36.89	.....	29.40	10.22		43.25	.....	2.23	2.11	16.38
11th ft.	15.65	21.56	.....	25.48	11.06		40.76	.....	1.34	4.87	16.64
12th ft.	14.30	35.84	.....	38.22	10.50		38.45	.....	2.62	1.44	10.85

humus and moisture, or in the presence of alkali salts, the activities of ammonifying organisms are seriously handicapped.

To discuss these more in detail we find, for example, in soil No. 1, derived from the mesa soil at Arlington Heights, Riverside, a strong ammonification, varying but little from the first foot down to the seventh, below which depth we find a sudden marked decrease in ammonia production, for the reason, doubtless, that from the sixth foot down to the twelfth we find a layer of hardpan which, owing to its poor aëration and poor water conditions, is unfit for the development of a vigorous bacterial flora. In other words, we find in this soil-column, through the ammonifying power of the various depths of soil, an expression of the vigor and numbers of bacteria present in these soil layers and also of the amounts of soluble nitrogen which can there be expected to be made available through the agency of soil organisms.

In soil No. 2, however, which represents a good, deep alluvial soil, we find a very vigorous ammonification from the first seven feet, and only slightly reduced ammonia production in the eighth and ninth feet, after which we find a large reduction of about 50 per cent in ammonia production for the other three feet. We have here, therefore, good vigorous ammonia production down

to the tenth foot and therefore an indication that in these soils there is constantly being made available nitrogen, if organic nitrogen be present from humus and other sources, for the needs of plants with deep root systems.

In soil No. 3, which is more sandy than the other alluvial soil described and which rapidly becomes coarser in texture as we descend into the lower layers, we find vigorous ammonification to obtain down to the fourth foot, below which we find a considerable decrease in ammonifying power, owing to the fact that in that coarse soil neither water nor humus, nor soluble minerals, are present in sufficient quantity to encourage bacterial development. Here, however, we find the general tendency for ammonifying organisms to penetrate to the greater depth quite plainly visible. The remarks made for soils 2 and 3 are just as truly applicable to the other alluvial soils from the same district represented by Nos. 4, 5, and 7. The marked production of ammonia, even in the twelfth foot of No. 7, is in accord with the fine physical and chemical condition of that soil to that depth and therefore deserves additional mention here.

As to other types of soils, the data in the table show plainly enough what a profound effect strong alkali salts (both black and white alkali salts among them) may exert on the ammonifying flora and their vigor. Here ammonification is indeed very feeble in the surface soil, becoming feebler as we go down until the eighth foot is reached, at which depth, as well as in the ninth foot, we find quite a marked increase in ammonia production. This is doubtless due to the fact that the total salt-content is at that depth much lower and therefore not so seriously affecting the activities of the organisms there contained. As for the desert soils, which never have contained much humus and very frequently contain too much alkali, it is natural to expect a rather feeble ammonifying power on the part of the soils. Table I shows that in this case the expected happens. In soil No. 9, for example, not only the lack of humus and moisture, but the very unfavorable physical condition, above referred to under the description of that soil, along with its salt-content, have so far affected the ammonifying power of that soil as to reduce it to a little over one-third of what the normal valley soils de-

scribed have exhibited. Moreover, it would seem that the salt-content in the lower layers of this soil, which increases as we go down, has very seriously checked the development of these organisms there and was probably assisted by the unfavorable physical condition mentioned. In soil No. 10, while the salt-content is only meager, we have a rather coarse, sandy soil with hardly any humus, which is therefore for that reason an unfavorable medium for the development of bacteria, to say nothing of the lack of moisture there and the great heat which these desert soils must absorb from the sun. We therefore have a very much smaller ammonifying power in the upper layers of the soil than exists even in soil No. 9, from Imperial, and then a very rapid decrease to almost no ammonifying power in the lower layers.

By a general survey of all of these data, it would certainly seem that we are justified in drawing the conclusion that ammonification and the ammonifying flora of soils are vigorous for several feet down in the arid region and are limited in their activities only by the presence of large amounts of salt or a lack of humus and moisture. Since, however, California soils, taken by and large, are deep, we have reason, from the facts above given, to suppose that the ammonifying power in most of these soils, which are not in any way "abnormal," is vigorous at great depths.

## NITRIFYING POWERS OF SOIL COLUMNS

By very many and perhaps by most plants, nitrate is the form of nitrogen taken up. It is therefore of importance not only to study ammonia formation in soils, but nitrate formation as well. In these investigations we have studied qualitatively and quantitatively the production of nitrites and nitrates in ammonium sulfate solution by soils from the different depths in every case. Here also, as in the ammonification work, 5 grams of soil were used to inoculate 50 c.c. of culture solution. The results obtained in this work are set forth in a qualitative manner, as to nitrate formation merely, in table II, since it is sufficient for the purpose of this preliminary paper to know to what depths in the soil nitrates are produced. Later publications, giving the more complete data of these investigations, will

give the quantitative results as they are given for ammonification in table I. The plus sign represents nitrate formation and the minus sign the absence thereof. The numbers of the soils are referred again to the descriptions above given.

TABLE II  
NITRIFICATION IN SOIL COLUMNS

Soil No.	1	2	3	4	5	6	7	8	9	10	11
1st ft.	+	+	+	+	+	+	+	—	—	—	+
2nd ft.	+	+	+	+	+	—	+	—	—	—	+
3rd ft.	+	+	+	+	+	—	+	—	—	—	+
4th ft.	+	+	+	+	+	—	+	—	—	—	+
5th ft.	+	+	+	+	+	—	—	—	—	—	+
6th ft.	trace	+	—	—	+	—	—	—	—	—	+
7th ft.	—	—	—	—	—	—	—	—	—	—	+
8th ft.	—	—	—	—	—	—	—	—	—	—	+
9th ft.	—	—	—	—	—		—		—	—	—
10th ft.	—	—		—	—		—		—	—	—
11th ft.	—	—		—	—		—		—	—	—
12th ft.	—	—		—	—		—		—	—	—

From the data in table II we see again a striking resemblance between nitrification in the soil depths and ammonification in the same. All the alluvial soils in particular show very uniform nitrate formation. The latter seems to be as much inhibited in soil No. 1 by the hardpan layer as is ammonification. In soils 2, 3, 4, and 5, as well as 7, we find a general tendency for nitrates to be formed in the first five feet and then an enfeebled power of nitrate formation, in some cases for one foot and in other cases a total loss of that power. In nearly all cases these run parallel with a similar decrease in ammonia formation, but it seems that nitrate formation is more seriously hampered by the conditions which curtail ammonia formation and particularly, it appears, by the lack of oxygen in the lower layers of the soil. This is an account, therefore, of the first case which has come to my notice of nitrification at any depths below two or three feet in the soil, and shows a marked difference in itself between soils formed and existing under humid and those formed and existing under arid conditions. Nitrogen therefore is available in



these soils, not only for those plants which are able to absorb ammonia nitrogen, but also for that larger class of normal plants which absorb their nitrogen in the nitrate form. It must be said here, however, that nitrate formation proceeded always more rapidly in soils from the first foot than in cultures prepared from the other depths. This may indicate a smaller number of nitrifying organisms in the lower layers of the soil or perhaps a less vigorous flora, but their activities are uniform from the second foot down to the last depth in which they show no activity as indicated in table II. In soil No. 6, which we find on analysis contained merely a trace of humus, that circumstance seems to have made the soil unfit for the development of the vigorous bacterial flora and is supported by the data in tables II and III. As to the nitrate formation in culture solutions by the inoculation with this Hanford soil, nitrates were produced only after a month's incubation and only in small quantities in the culture prepared from the upper foot of soil, whereas all other surface soils, when inoculated into solutions with the exception of those which show no nitrification at all, showed nitrate formation before the end of two weeks. In culture solutions from soil No. 6, kept about three months and prepared from the lower layers of soil, no nitrates were ever to be found. The depressing effect of alkali on the bacterial flora, as well as the inhibiting effect of a lack of humus, moisture, and the proper physical condition, are again exemplified in table II in soils 8, 9, and 10, as they were for the same soils in table I referring to ammonification. Even after one month's incubation, not one of these soil-samples showed any nitrate formation, whether the culture was prepared with the soil from the upper layers or from the lower. There seems to be a total absence of nitrifying bacteria of one kind or another.

The best example of the penetration of nitrifying bacteria to great depths was obtained in soil No. 11, a fine alluvial loam from Hayward, where nitrate formation was obtained down to the ninth foot in the soil. In this case also there was, besides a mere formation of nitrates, as shown by a qualitative test, an actually vigorous nitrate formation in the lower layers as well as in the upper layers of the soil. It would seem again here

therefore, in general, that where soils in the arid region are supplied with a moderate amount of humus, with the proper texture and chemical constitution, as well as freedom from alkali, all of which is true of the large majority of our soils, nitrification as well as ammonification is found to obtain vigorously in the lower layers of the soil for four feet at least, and in some cases to six and to nine foot depths.

### NITROGEN FIXATION IN SOIL COLUMNS

The next point of interest to determine in these soil-column investigations from the bacteriological standpoint was to show whether or not the supply of nitrogen, at the disposal of the ammonia-forming and nitrate-forming organisms, which we have found developed to such great depths, and enabling roots to have a soluble nitrogen supply there, was provided merely by the humus content of the soil at those depths and produced from decaying roots, or carried down from the upper layers; or, was that nitrogen supply in part a new one obtained directly from the atmosphere by nitrogen-fixing bacteria. If such were the case, we should, of course, have enormous quantities of nitrogen fixed per acre, since the fixation would not be limited to the upper foot of soil. Accordingly, experiments were inaugurated to obtain the facts which exist with reference to this matter.

Here the necessary mannite solution was inoculated with five grams of soil in each case, and a culture prepared from every foot in depth in the case of every soil. Table III shows in tabular form the results obtained, which are set forth qualitatively. The numbers at the heads of the columns refer again to the numbers used in the description of soils, and one plus sign is intended to show the presence of *Azotobacter*, two of a fairly vigorous development of these organisms, and three of a very vigorous development. In this qualitative way, therefore, nitrogen fixation has been judged by the development of *Azotobacter* as a criterion. It may justly be argued against this that other organisms are capable of fixing nitrogen and that the quantitative figures would be preferable to the qualitative one showing merely the presence of *Azotobacter*. While this argument may in part be true, it appears from my results, which

show quantitative as well as qualitative figures in these as well as other experiments, that in the absence of *Azotobacter* only very slight fixations of nitrogen or none are obtained.

From the results set forth in table III, it appears that only one soil of the eleven tested shows the presence of *Azotobacter* as deep down as the fourth foot and six others show the presence of these organisms in the third foot. Most of them, however, show the presence of *Azotobacter* in vigorous form only in the first two feet. It is therefore not sufficient, evidently, for

TABLE III  
NITROGEN FIXATION IN SOIL COLUMNS

Soil No.	1	2	3	4	5	6	7	8	9	10	11
1st ft.	+++	+++	+++	+++	+++	—	+++	—	—	—	++
2nd ft.	++	+++	+++	+++	+++	—	+++	—	—	—	++
3rd ft.	+	—	++	+++	+++	—	++	—	—	—	++
4th ft.	—	—	—	—	+++	—	—	—	—	—	+
5th ft.	—	—	—	—	—	—	—	—	—	—	—
6th ft.	—	—	—	—	—	—	—	—	—	—	—
7th ft.	—	—	—	—	—	—	—	—	—	—	—
8th ft.	—	—	—	—	—	—	—	—	—	—	—
9th ft.	—	—	—	—	—	—	—	—	—	—	—
10th ft.	—	—	—	—	—	—	—	—	—	—	—
11th ft.	—	—	—	—	—	—	—	—	—	—	—
12th ft.	—	—	—	—	—	—	—	—	—	—	—

soils to be chemically and physically as favorably constituted as these soils are for ammonification and nitrification to encourage the deeper penetration of *Azotobacter*. As is well known, these organisms are extremely sensitive to a lack of oxygen and it would appear that this circumstance regulates and controls the penetration of *Azotobacter* organisms as above portrayed. I think in addition, however, it may fairly be argued that the presence of *Azotobacter* in more than half of these soils in the third foot is, in itself, a favorable indication of the nature of the soils in question. It is of interest also that, in soil No. 5, *Azotobacter* organisms, with the nitrogen-fixing power as vigorous as those above, were found in the fourth foot. This, so far as the writer is

aware, constitutes the only published case of even this extent of penetration of *Azotobacter* organisms. It has been reported to me, however, that *Azobacter* organisms have been found in the twelfth foot of soil in some of the very favorably constituted loess soils of Nebraska. The question put in the introduction to this subject of nitrogen fixation is therefore answered in the negative. For the greater depths, at any rate, in which ammonification manifestly is vigorous in our soils, *Azotobacter* organisms do not penetrate and are not the source of the supply of nitrogen which can be transformed at those depths by the ammonifying organisms or by the nitrifying organisms. The nitrogen supply of these, therefore, in the lower layers of the soil must be the humus produced from the decaying roots at those depths, or the humus brought down in solution from the upper layers of the soil.

As regards soil No. 6 we have here again a total absence of *Azotobacter* organisms, possibly due in part, at least, if not wholly, to the absence of any but very small amounts of humus, by which I have already tried to explain the feeble nitrification only in the first foot of this same soil. The same remarks also which were made above, with reference to soil Nos. 8, 9, and 10, as regards their ammonifying and particularly their nitrifying power, apply again in the case of their nitrogen-fixing power. No *Azotobacter* organisms and no fixation of nitrogen were ever observed in any of these soils, no matter from what depth of soil the cultures were prepared.

In justice to this subject it must further be stated here that the comparatively slight penetration of *Azotobacter* organisms in our soils may be due to factors other than merely a lack of a plentiful supply of oxygen. There is evidently some other circumstance which controls the presence or absence in many of our soils of *Azotobacter* organisms and that may also limit the depth to which these organisms may penetrate. Just what this factor may be is not at present clear to the writer, but the fact remains that frequently soils with a good chemical and physical constitution and producing good crops, will yet show no *Azotobacter* organisms.

## GENERAL DISCUSSION

As I have already pointed out in an earlier publication,<sup>2</sup> the slow formation of clay substances in soils of the arid region, owing to the peculiar climatic conditions there obtaining, is doubtless responsible for a much greater degree of aëration in soils because of the larger volume of pore spaces made possible through a lack of large quantities of cementing substances. Thus when soils first begin to form from disintegrating rock we have much more complete aëration with an encouragement for bacteria, probably the earliest inhabitants of the soil, to penetrate to greater depths. Such penetration on the part of bacteria is invariably accompanied by the production of more favorable physical and chemical conditions in the soil for the roots of plants. These in their turn, through physical and chemical changes which they bring about in the soil in their search for water and food, make better conditions for a deeper penetration of bacteria and so through mutual aid the latter and the higher plants are able, under our arid climatic conditions, to make the deeper layers of soil a more congenial medium for each other. The changes thus brought about result in a more uniform texture of soils at great depths, uniformity of chemical composition, including humus content, in all the soil layers, and a much closer approximation of the bacterial flora in the lower soil layers to those of the upper layers than can be found in the average soils of the humid region, where climatic conditions are unfavorable to good aëration, because tendencies opposite to those above described for our soils are in operation. An estimate of the biological condition of our deep soils was thus similarly made by Hilgard on *à priori* considerations and the investigations above recorded serve, in general, to confirm his surmise.

Viewing the subject in its entirety, we find that the organisms forming ammonia in soils penetrate to greater depths than the nitrifying or nitrogen-fixing bacteria studied. While ammonification is usually most vigorous in the surface, four to six feet, it is none the less very pronounced in the lower layers from six to ten feet in depth in all of our normal deep soils. Hardpan,

<sup>2</sup> Lipman, C. B., New Facts about Bacteria of California Soils, Science N. S., June 11, 1909.

alkali, and a lack of humus and moisture decrease the ammonifying powers of our soils or are not favorable to the development of vigorous ammonifying flora, but their effects are just as pronounced in the upper layers of these abnormal soils as in the lower layers which, therefore, cannot be fairly compared with our deep average soils as to bacterial content. To what a serious extent alkali salts may affect ammonification has been shown by me in a recent paper.<sup>3</sup> That the humus content alone may profoundly affect the number and vigor of bacteria is well exemplified in both soils No. 6 and 10, where all other conditions but the humus content are favorable and where both the number and physiological efficiency of the organisms is small.

It would therefore seem, in brief, that ammonification is vigorously active in the lower soil layers in soils of the arid region where humus is present and hardpan and alkali are absent. Since these conditions are complied with in the average of our cropped soils, the opinion is justified that the deep penetration of bacteria is a distinctive characteristic of soils in arid regions which results from much better aëration, as a starting point, than can be attained in soils of the humid region. The experimental data above given amply confirm this opinion and help to explain why deep plowing is not only harmless in our soils but directly beneficial, and why three or four feet of upper soil may be removed in grading, and alfalfa and fruit trees may be grown on the newly uncovered subsoil without difficulty, a feat which cannot be accomplished on soils of the humid regions.

As for nitrification my data present again features of striking interest. They go to prove that nitrate formation, like ammonification, goes on at much greater depths in soils of arid than in soils of the humid region, and thus render distinctly sectional the observations of Dyer<sup>4</sup> on this subject, and makes them applicable only to soils of the humid region. While the nitrifying organisms are doubtless more susceptible to a lack of oxygen than the ammonifying bacteria, the differences obtained above between the two groups of organisms, so far as soil fertility is concerned, are rather those of degree than of kind. The same relationships

<sup>4</sup> Bulletin 106, p. 55, O. E. S., U. S. D. A.

<sup>3</sup> Centrallblatt für Bakt., 2 Abt., vol. 32, p. 58.

displayed by the ammonifying bacteria toward hardpan, alkali, and a lack of humus and moisture, hold in a more exaggerated way as regards the nitrifying organisms. More specifically, the writer has also shown<sup>5</sup> the distinct effects of each of the alkali salts on nitrifying bacteria in work quite recently completed. It would appear in general, however, that in our deep soils, a supply of nitrate as well as of ammonia is at the disposal of plants for a depth of five or six feet. As regards the nitrogen-fixing powers of soils of the arid region, my results show plainly that they do not differ strikingly from those of soils in the humid regions, if the presence and vigor of *Azotobacter* organisms be taken as a criterion. While it is true that in one or two cases *Azotobacter* organisms were found in our soil-columns below the depth at which they occur elsewhere, and perhaps at a slightly greater depth in all soils in which they were found, I feel loath to believe that these are expressions of a rule for soils of the arid region. Other observations indeed lead me to believe that *Azotobacter* development has not gone so far in our soils as it has in soils of other regions. For example, I have studied many soils in California with a favorable physical and chemical constitution which were absolutely devoid of *Azotobacter* organisms. If therefore the results set forth above, with reference to nitrogen fixation, are to be considered representative, the nitrogen supply in the lower layers of the soil must be replenished in this region as well as in the humid region, not from direct fixation by *Azotobacter*, but from the nitrogen of the upper soil layers.

With reference to these investigations in general, one or two additional points need more than passing consideration. First, as to the method of collecting the soil-samples for examination, it appears to the writer that every possible precaution was used to prevent contamination and it would be difficult to devise a method which takes into consideration and avoids more of the avenues of contamination by which any results might be vitiated. Moreover, I find strong confirmation of this belief in the facts brought out in the data above given, viz., that any abnormality in the soil was sure to be reflected in the results obtained<sup>6</sup> with cultures prepared from that abnormal soil. Thus hardpan layers

<sup>5</sup> *Cent. für Bakt.*, 2 Abt., vol. 33, p. 305.

never gave evidence of vigorous bacteria, nor did alkali soils or soils devoid of humus.

Secondly, the writer desires to anticipate criticism on the method used in culturing the organisms of the various soil samples, viz., a modified Remy solution method. No one is more ready than I am to admit the just criticism made of the solution-culture methods in soil bacteriology. Indeed I believe that I was one of the first to put into practice on a large scale the *direct* soil-culture method in the laboratory. But when problems of the nature involved in these investigations must be attacked, regard must be had for the chances of contamination in the method employed, and for the feasibility of obtaining, uncontaminated, large volumes of soil for use in these experiments. When these were considered from all points of view, only one feasible and reliable method of culturing the soils seemed available and that was the solution method. The difficulties, practically insurmountable, which must arise with any other method, when such work is carried out on a large scale as it must of necessity be, can be fully appreciated by those who have ever attempted it. The gratifying results obtained in this work, however, seem to me a further justification of the methods employed.

It seems of particular moment now, to call the attention of soil bacteriologists in particular, and soil scientists in general, to the important field explored in these investigations and the striking results obtained therefrom, not only because it represents a new field of research, but because it emphasizes more strongly than ever the radical differences which obtain between soils of the humid and arid regions. It also helps to explain the extraordinary appearance of our subsoils (if subsoils they be) and the marvellous root developments of which plants under our climatic conditions are capable. While these studies have not yet departed from the realms of the preliminary, they are replete with facts which are already of considerable practical and scientific significance and which are doubtless destined to become more so as time progresses. As a part especially of a comprehensive soil study they are invested with unusual importance and may help to solve problems now perplexing and difficult to study.



## CONCLUSIONS

Investigations of the distribution and activities of bacteria in soils of the arid region show:

1. That samples of soil for studying the flora of each layer of soil can best be obtained from a hole twelve feet in depth with at least one vertical wall, the latter when sterilized being sampled.

2. That tin tubes ten inches long and about one inch in diameter closed at one end and cotton-stoppered are best for collecting the samples.

3. That the solution method for studying the soils, despite its many drawbacks, is the most feasible one to employ.

4. That soils of the arid region at all depths studied show ammonifying powers which, however, are generally most vigorous in the first six or eight feet. In one case ammonification was noted in soil from a depth of fifteen feet, or adjoining the water-table.

5. That nitrification is found commonly down to a depth of five to six feet in soils of the arid region. In one case soil from the eight-foot depth showed a vigorous nitrifying power.

6. That nitrogen fixation through *Azotobacter* does not go on below two feet in the soil usually, but has been found in some soils at three feet and in one soil down to four feet. Many soils in the arid region, otherwise favorably constituted, do not contain *Azotobacter* organisms.

7. That from the point of view of ammonification and nitrification soils in the arid region differ markedly from those in the humid region when the lower layers of soil are considered. The difference is not marked as regards nitrogen fixation.

8. The results above recorded help to explain the favorable physical and chemical constitution of our soil and also the deep rooting of plants so characteristic of the arid regions.

*Transmitted April 8, 1912.*





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STUDIES ON THE PHENOLDISULPHONIC  
ACID METHOD FOR DETERMINING  
NITRATES IN SOILS

BY

C. B. LIPMAN and L. T. SHARP

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ACID METHOD FOR DETERMINING  
NITRATES IN SOILS

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C. B. LIPMAN AND L. T. SHARP

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Despite the fact that some careful research has been carried out on the colorimetric method for determining nitrates, many factors concerned with it have not been studied, and the somewhat uncertain nature of the method makes it imperative to control, so far as possible, every factor which may interfere with the accurate analysis of nitrate-containing material. These statements apply particularly to the analysis of soils for nitrates and the authors therefore deem the subjoined data, derived from a thorough investigation, deserving of the attention of every soil chemist.

Among the interfering factors in the phenoldisulphonic acid method which have been either studied inadequately or not at all, are the effects of salts, the effects of agents employed to precipitate the clay and organic matter, and the effects of decolorizing agents. Cognizance must be taken of all of these factors by the chemist in the determination of nitrates and in the arrangement and interpretation of results. The importance of salt effects and their significance in this connection are emphasized by the fact that many soils, and particularly those of arid and semiarid regions, may frequently be found to a greater or less degree impregnated with one or more of the so-called "alkali salts," together with which, it often happens indeed, consider-

able quantities of nitrates are to be found. So far as the clay-coagulating substances are concerned, it has always been a common practice in soil work to employ varying amounts of a saturated solution of alum to obtain a clear soil solution, and more recently it has been proposed by investigators who have studied the method under discussion to use aluminum cream for the purpose in place of alum. For decolorizing solutions both aluminum cream and bone black have been used. The methods for both clay coagulation and decolorization are obviously essential in most soil work, since ordinary filtration, without the use of such agents, can rarely be depended on to yield a clear, colorless soil solution, even if time be no object. The employment of the Pasteur Chamberland filter to remove clay has been found by direct investigation to involve well-defined losses of nitrates.

It is not our purpose here to enter into a lengthy review of other investigations bearing on the subject in hand, but into a brief discussion of the more important ones which show the questions still remaining unsolved or bring out certain results with which ours do not agree.

In 1894 Gill<sup>1</sup> carried out a series of painstaking investigations which, briefly, indicate (1) that for purposes of accuracy the phenoldisulphonic acid employed in the nitrate determination must be carefully prepared to insure a uniform compound for use as a standard; (2) that chlorine induces losses of nitric acid both when the solution containing nitrate is evaporated on the water bath and when the residue is treated with the reagent; (3) that  $\text{Na}_2\text{CO}_3$  added to the nitrate-containing solution to prevent escape of nitric acid during evaporation induces losses of nitrates varying in quantity from four to six per cent; (4) that alumina may be used to precipitate colloidal material for obtaining a clear solution; (5) that silver sulphate, if free from nitrate, may be employed to precipitate chlorine, thus removing an important interfering agent.

More recently Chamot and his coworkers<sup>2</sup> have prosecuted an even more thoroughgoing investigation than the preceding, in which the most emphasis has been placed, however, on the mode of preparation of the tripotassium salt of nitrophenoldisulphonic

<sup>1</sup> *Jour. Am. Chem. Soc.* vol. 16, p. 122. 1894.

acid used as the reagent. Their results indicate (1) that in order to obtain the phenoldisulphonic acid free from the mono and tri-phenolsulphonic acid a careful digestion of the phenol and sulphuric acid under certain constant conditions must be assured; (2) that the mono and tri-phenolsulphonic acids introduce other colors which interfere with the readings in the colorimeter; (3) that the tri-potassium salt of nitrophenoldisulphonic acid gives the characteristic color employed in the determination and should always be used as a standard; (4) that heating the dry residue of nitrates even for several hours on the water bath occasions no losses; (5) that aluminum cream is the best precipitating agent for organic matter of several used and occasions no losses of nitrates; (6) that 2 c.c. of the phenoldisulphonic acid should be used in uniform amounts in all determinations; (7) that KOH was to be preferred to NaOH and  $\text{NH}_4\text{OH}$ , as the alkali employed; (8) that chlorides induced losses of nitrates; (9) that carbonates and organic matter did likewise; (10) that temperature, concentration, and length of exposure to reagent greatly affect results; and (11) that there have been other minor effects of iron, magnesium, and nitrites.

Reference must also be made here to the brief investigation of Stewart and Greaves<sup>2</sup> pertaining to the effect of chlorine in determining nitrates in soils, both because the work is recent and because it is the only one published which is derived from researches on soils. This investigation and those above reviewed cover most completely the questions involved and reference will be made in the discussion of our experimental work below to those questionable points which were considered settled but which our work shows were far from being so.

#### THE INTERFERENCE OF SALTS WITH THE NITRATE DETERMINATION

As has been above indicated the salt accumulations which occur in the soils of California, Nevada, Utah, and other arid or semi-arid regions frequently contain considerable quantities of nitrates and the determination of the latter in the presence of the "alkali salts" is, as has been found, frequently attended

<sup>2</sup> *Ibid.*, vols. 21, p. 922; 32, p. 630; 33, p. 366

<sup>3</sup> *Ibid.*, vol. 32, p. 756.

with losses of nitric acid. While all the investigations above reviewed have pointed out the interference of chlorine and chlorides with the nitrate determination, and while some of them have also considered the losses occurring through the use of  $\text{Na}_2\text{CO}_3$ , no mention is made of the effects of the most common and widely spread of the alkali salts,  $\text{Na}_2\text{SO}_4$ , or Glauber salt. It seems further to have been taken for granted that  $\text{Na}_2\text{CO}_3$  and  $\text{Na}_2\text{SO}_4$  should, for obvious reasons, have the same effects on the nitrate determination by the phenoldisulphonic acid method. Our results do not, however, bear out this opinion. Under this head were also studied the effects of the kation as well as the anion of salts on the same determination.

Varying quantities of the salts tested were here added to the same amounts of nitrates in solution, and uniform quantities of salts were also tested as to their effects on varying quantities of nitrates. Everyone of the following tables gives the effects of one of the salts tested in accordance with the scheme above indicated and in some cases also shows how the nitrate determination is affected by varying the quantities of both the nitrates and other salts. The residue containing the salts and the nitrates was treated with 2 c.c of phenoldisulphonic acid thoroughly stirred for about two or three minutes, 25 c.c. of nitrate-free distilled water was added, and then strong ammonia drop by drop until the odor of ammonia persisted and the color was permanent. The solution was then diluted as necessary and compared in the Sargent-Kennicott colorimeter with a standard solution similarly and always freshly prepared, whose strength was in every case carefully tested. The results of these experiments are given in the following tables.

TABLE I  
EFFECTS OF NaCl

	NaCl added mgs.	N. added as nitrate mgs.	N. found as nitrate mgs.
Uniform quantities	.25	.050	.045
KNO <sub>3</sub> —varying	.50	.050	.041
amounts NaCl	1.00	.050	.035
	2.50	.050	.026
Varying quantities	1.00	.050	.038
KNO <sub>3</sub> -uniform	1.00	.100	.070
amounts NaCl	1.00	.250	.215
	1.00	.500	.460
	1.00	1.000	.940
	1.00	2.500	2.300
Varying quantities of	.25	.050	.046
both KNO <sub>3</sub> and NaCl	.50	.100	.078
	1.00	.250	.230
	1.50	.500	.460
	2.00	1.000	.900
	2.50	2.500	2.300
Uniform amounts KNO <sub>3</sub> -	.01	.050	.051
small amounts NaCl	.05	.050	.051
	.10	.050	.049
Color blanks	.00	.050	.050
on both salts	2.50	.000	.000

TABLE II  
EFFECTS OF Na<sub>2</sub>SO<sub>4</sub>

	Na <sub>2</sub> SO <sub>4</sub> added mgs.	N. added as nitrate mgs.	N. found as nitrate mgs.
Amounts of nitrate	1.000	.0500	.0480
uniform and sulfate	5.000	.0500	.0420
varying	10.000	.0500	.0400
	20.000	.0500	.0280
	30.000	.0500	.0270
Amounts of sulfate	15.000	.1500	.1420
uniform and nitrate	15.000	.5000	.4950
varying	15.000	1.0000	.9000
	15.000	2.0000	1.9500
	15.000	3.0000	2.8500
Amounts of both salts	1.000	.1500	.1420
varying	5.000	.5000	.4800
	10.000	1.0000	.9200
	20.000	2.0000	1.9300

TABLE III  
EFFECTS OF  $\text{Na}_2\text{CO}_3$

	$\text{Na}_2\text{CO}_3$ added mgs.	N. added as nitrate mgs.	N. found as nitrate mgs.
Amounts of nitrate	1.00	.1000	.0995
uniform and carbonate	2.50	.1000	.1040
varying	5.00	.1000	.1010
	10.00	.1000	.1010
	20.00	.1000	.1020
	30.00	.1000	.1020
Amounts of carbonate	10.00	.1000	.1000
uniform and nitrate	10.00	.2000	.2100
varying	10.00	.5000	.5000
	10.00	1.5000	1.4900
Amounts of both salts	1.00	.1000	.1010
varying	5.00	.2000	.1970
	10.00	.5000	.5050
	20.00	.5000	.5100
	30.00	1.5000	1.4900

The results set forth in tables I, II, and III leave no room for doubt as to the effects of "alkali" salts on the nitrate determination by the colorimetric method. Both  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$  induce large losses of nitrate, and especially is this true of  $\text{NaCl}$ , which may be responsible for losses equivalent to forty-five per cent and more of the total nitrate present as indicated in Table I. While  $\text{Na}_2\text{SO}_4$  induces smaller absolute losses than  $\text{NaCl}$ , they are none the less marked, and where large amounts of the sulfate are present very considerable losses of nitrate occur.

Perhaps the most striking feature of the foregoing results is what appeals to one at first sight as the singular difference in the behavior of  $\text{Na}_2\text{SO}_4$  and  $\text{Na}_2\text{CO}_3$ . Whereas the former is always responsible for losses in the determination of nitrates, the latter is the only one of the salts tested which has no effect and the presence of which in a long series of tests has never, except in one case, decreased the amount of nitrate present as shown by the colorimeter readings. It was naturally assumed that  $\text{Na}_2\text{CO}_3$ , after the addition of the phenoldisulphonic acid, would be converted in the presence of an excess of sulphuric acid into  $\text{Na}_2\text{SO}_4$  and should therefore show the same decreases in the nitrate content as the latter salt. To clear up these rather puzzling facts, as above given, we decided to run a special series of experiments based on a suspicion which we had as to the

nature of the action of the salts in question. The results of these experiments, which will be given below, make entirely clear what seemed at first quite puzzling.

In further general discussion of the tables above given, it must be added that the decreases in the nitrate content of the solutions tested as induced by the presence of salts never occurred in accordance with any definite law, the losses at times being greater with smaller amounts of salts than with larger amounts, the amounts of nitrates being constant. On the other hand, with a given amount of nitrates not exceeding one-tenth of a milligram the salts seemed always to induce larger percentage losses than they did in the case of the larger amounts of nitrates. Our results not only give good opportunity for a comparison of the effects of varying quantities of salts on the same nitrate content, but point out all the relationships between the salts and nitrates where first the former, then the latter, and finally both, are varied. There are two other points, also, which they would not seem to confirm; indeed they give entirely different evidence on these than was obtained by other investigators. The first is that small amounts of NaCl *do not* induce losses of nitrates, as claimed by Stewart and Greaves, and Table I indicates that amounts of NaCl below .1 milligram do not occasion any losses. The other point of difference between our results and those of the others mentioned is that  $\text{Na}_2\text{CO}_3$  does not decrease the amounts of nitrates, no matter to what extent it is used, as shown in Table III. This is in entire disagreement with the results of Gill and Chamot and his coworkers, who claimed that  $\text{Na}_2\text{CO}_3$  and other carbonates induced losses of nitrates, in the determination outlined. It must also be added here that the effects of  $\text{Na}_2\text{SO}_4$  as given in Table II constitute the first published results, so far as we are aware, on the effects of Glauber salt on the nitrate determination, and they have indeed been indirectly responsible for the discovery of one or two other points of interest which will be discussed below.

The results above given indicate the effects of each of the salts taken singly on the nitrate determination. To make the data more complete it was thought desirable to test various mixtures of the same salts and note their effects. Table IV gives the results obtained.



TABLE IV

## EFFECTS OF MIXED ALKALI SALTS

$\text{Na}_2\text{CO}_3$ mgs.	$\text{Na}_2\text{SO}_4$ mgs.	$\text{NaCl}$ mgs.	N. added as nitrate mgs.	N. found as nitrate mgs.
1	1	1	.1000	.055
5	5	5	.1000	.026
10	10	10	.1000	.021
20	20	20	.1000	.017
1	1	....	.1000	.082
5	5	....	.1000	.081
10	10	....	.1000	.075
20	20	....	.1000	.077
1	....	1	.1000	.061
5	....	5	.1000	.060
10	....	10	.1000	.055
20	....	20	.1000	.028
....	1	1	.1000	.050
....	5	5	.1000	.042
....	10	10	.1000	.033
....	20	20	.1000	.030
10	10	10	.2000	.086
10	10	10	.5000	.125
10	10	10	1.000	.360
10	10	10	2.000	1.140

The same marked losses in nitrates occur here as where the salts are employed singly.  $\text{NaCl}$  seems to be responsible again for the greatest losses,  $\text{Na}_2\text{SO}_4$  is next in order, and  $\text{Na}_2\text{CO}_3$  seems to have little or no effect. Since these salts occur together in alkali soils, however, the results in Table IV possess considerable significance and interest, especially since they point out what enormous losses of nitrates occur where such large amounts as ten milligrams of each of the salts are added to the nitrate-containing solution.

#### THE INTERFERENCE OF PRECIPITANTS OF CLAY AND ORGANIC MATTER ON THE NITRATE DETERMINATION

It is very singular that analytical chemists have for so long a time been employing such materials as saturated alum solutions, aluminum cream, and bone black for precipitating clay and organic matter in obtaining the soil solution to be used for nitrate determinations without ever having attempted to ascer-

tain if such materials in any way affect the accuracy of the determination. Indeed Chamot and his coworkers have recommended the use of aluminum cream for removing suspended material from the solution, and claim to have had very satisfactory results in the use of that material. Our experiments in this series were intended to clear up this question and the following results show very strikingly that none of the materials mentioned may be employed in the nitrate determinations without incurring very serious losses. Table V gives results obtained in the use of potash alum, and Table VI gives results obtained in the use of bone black and aluminum cream.

TABLE V

EFFECTS OF  $K_2Al_2(SO_4)_4$ 

	$K_2Al_2(SO_4)_4$ added mgs.	N. added as nitrate mgs.	N. found as nitrate mgs.
Amounts of nitrate	5.00	.050	.040
uniform and alum	12.50	.050	.036
varying	25.00	.050	.033
	50.00	.050	.031
	100.00	.050	.034
	150.00	.050	.040
Amounts of alum uniform	45.00	.050	.035
and nitrate varying	45.00	.100	.075
	45.00	.250	.168
	45.00	.500	.345
	45.00	1.000	.675
	45.00	2.500	1.800
Amounts of both	5.00	.050	.040
salts varying	12.50	.100	.074
	25.00	.250	.175
	50.00	.500	.335
	100.00	1.000	.690
	150.00	2.500	1.850
Color blanks on	.00	.050	.049
both salts	.00	.500	.480
	100.00	.....	.....

**TABLE VI**  
**EFFECTS OF ALUMINUM CREAM AND BONE BLACK**

	N. added as nitrate mgs.	N. found as nitrate mgs.
Sufficient aluminum cream to clear solution. Five minutes exposure	.5000 1.000 2.000	.254 .648 1.460
Twice the amount of aluminum cream used above. Exposed one and one-half hours	.5000 1.0000 2.0000	.100 .300 1.180
Sufficient bone black to clear and decolorize solution	1.0000 2.5000 5.0000	.135 .650 2.200

The data in Tables V and VI are clearly very striking. The enormous losses of nitrates sustained through the use of a saturated solution of alum, varying quantities of aluminum cream and bone black, make these substances entirely unfit for use as precipitants for clay, or organic matter, or both, when nitrates are to be determined. While bone black occasions the largest losses, and potash alum the smallest, of any of the substances above described, the losses of nitrates brought about through the use of all the precipitants are too great to permit of their continuance in a method for nitrate determinations which is none too accurate under the best of conditions. It is therefore evident that nitrates are lost not merely through the loss of nitric acid, as is the case where salts are used, but that there is a loss of nitrates mechanically through adsorption on the part of the colloidal material of the precipitant, as must be the case where such substances as aluminum cream and bone black are used. The large amounts of colloids possessed by these substances, with the accompanying large surface areas, evidently prevent some of the nitrate in solution from going through the filter.

On casting about for a method to precipitate clay or organic matter, we first tried the Briggs filter pump, but found that open to two objections. First, the losses of nitrates through what we look upon as adsorption on the part of the clay filter, though not very large, were nearly equal to those induced by small amounts of sulfates. Second, while the filter pump yields a clear solution, it does not serve to decolorize solutions. After several fur-

ther attempts to find a coagulating and decolorizing agent which might promise well for this method, it struck us that quicklime, being the best coagulating material for clay, might perhaps also serve to remove organic matter and yet might not decrease seriously the amount of nitrates in the solution to be tested. Accordingly, tests were carried out by adding lime to solutions containing known amounts of nitrates, to soils containing known amounts of nitrates and to soils with unknown amounts of nitrates, in which latter a comparison was also especially made between lime and aluminum cream. We found in these experiments that the losses of nitrate through the use of lime were not only very small or negligible, but that the action of lime in precipitating both clay and organic matter was equal to or better than that of the best of the coagulating and decolorizing agents. Its coagulating action on clay has of course always been recognized in soil physics. The results of the experiments are given in Table VII.

TABLE VII  
EFFECTS OF LIME

A—Solutions of known nitrate content

CaO present grms.	N. added as nitrate mgs.	N. found as nitrate mgs.
1	1.0000	1.0150
3	1.0000	.9800
5	1.0000	.9550
3	5.0000	4.6500

B—Soils of known nitrate content

	CaO present grms.	N. present as nitrate mgs.	N. found as nitrate mgs.
Lime ground with soil and water	2	3.280	3.150
Lime added to muddy suspension	2	3.280	3.200

C—Comparison of lime and aluminum cream on soil of unknown nitrate content

	CaO present grms.	N. found as nitrate mgs.
Lime ground with soil and water	2	1.210
Lime added to muddy suspension	2	1.225
Sufficient aluminum cream added to clear solution	..	.800

It would seem from these results therefore that lime can yield a clear, colorless solution without decreasing the quantity of nitrates present in the solution appreciably, and that it is therefore the only one of the coagulating agents above tested which can be safely used in the work. We commend it to soil chemists and others making nitrogen determination under similar conditions. Only where very large quantities of lime are employed, and they are not necessary, have we found definite losses of nitrates. We find that 2 grams of  $\text{CaO}$  is sufficient to coagulate the clay in 100 grams of loam soil and to remove whatever color may be present at the same time.

While lime has been used by some chemists in accordance with the method above outlined, its use has by no means been general and no data prior to this existed with reference to its effects on the nitrate determination. J. G. Lipman and P. E. Brown give directions in their laboratory manual on Soil Bacteriology for the use of 2 grams of lime to precipitate the clay in the 100 gram samples of soil used in nitrification experiments, but we have never seen any published statements beyond that as to the advisability or feasibility of employing lime. It is certainly surprising that those who have tested the method for nitrate determination should not have tried and urged the use of lime as a substitute for alum or aluminum cream.

#### OTHER EXPERIMENTS ON SALT EFFECTS

It appeared interesting, when the results in Tables I, II, and III were obtained, to ascertain if the kation as well as the anion of salts was responsible for losses of nitrates. Accordingly a series of experiments was instituted in which the effects of  $\text{NaCl}$ ,  $\text{KCl}$ , and  $\text{MgCl}_2$  could be compared. The following results were obtained.

TABLE VIII  
EFFECTS OF IONS

KCl mgs.	MgCl <sub>2</sub> mgs.	NaCl mgs.	N. added as nitrate mgs.	N. found as nitrate mgs.
1	....	....	.1000	.070
5	....	....	.1000	.063
10	....	....	.1000	.055
20	....	....	.1000	.050
....	1	....	.1000	.057
....	5	....	.1000	.028
....	10	....	.1000	.016
....	20	....	.1000	.011
....	....	1	.1000	.065
....	....	5	.1000	.043
....	....	10	.1000	.035
....	....	20	.1000	.038

It is evident from Table VIII that the chlorine and not the base is the interfering element, and while the amounts of chlorine were not so proportioned as to be equivalent in the case of the two monovalent bases, the effect is clearly seen of the smallest and the largest amounts of chlorine present in the salts, which can be calculated from the molecular weights. The negative ion therefore seems to be the active agent in setting free nitric acid, but the decreases, depending as they do on other conditions such as evaporation on the water bath and length of exposure, do not take place in accordance with any definite law.

The last phase of the salt effects studied was that above referred to in the discussion of Tables I, II, and III, namely, the reason for differences in the action of  $\text{Na}_2\text{CO}_3$  and  $\text{Na}_2\text{SO}_4$  on nitrate-containing material. Since it was evident that  $\text{Na}_2\text{CO}_3$  should react similarly to  $\text{Na}_2\text{SO}_4$  when the phenoldisulphonic acid was added to the dried residue to be analyzed, we suspected that the losses occurring when  $\text{Na}_2\text{SO}_4$  was employed came about on the water bath in evaporating the solution, under which conditions only, according to our work, could there have been a difference in the action of the two salts.

The results given in the following table prove that our suspicions were well founded. In this series the dry salts were thoroughly mixed with the nitrate-containing residue obtained by evaporating standard nitrate solutions, and then the phenoldisulphonic acid reagent was added. NaCl was similarly tested.

TABLE IX  
EFFECTS OF DRY MIXING OF NITRATES AND SALTS

$\text{Na}_2\text{CO}_3$ mgs.	$\text{Na}_2\text{SO}_4$ mgs.	$\text{NaCl}$ mgs.	N. added as nitrate mgs.	N. found as nitrate mgs.
50	....	....	.1000	.097
100	....	....	.1000	.102
....	50	....	.1000	.103
....	100	....	.1000	.102
....	....	50	.1000	.080
....	....	100	.1000	.062

The data in Table IX make it quite clear that the losses due to  $\text{Na}_2\text{SO}_4$  occur only when the latter salt is present in solution with nitrates and the solution is evaporated on the steam bath. When, however, the salt is mixed dry with the dry nitrate no losses of the latter occur any more than they do when  $\text{Na}_2\text{CO}_3$  is added. The same is not true, however, of  $\text{NaCl}$ , as is shown in the last table. That salt causes losses of nitrates during both the evaporation on the steam bath and the reaction setting chlorine free in the treatment of the dry residue with phenoldisulphonic acid. This latter fact is a confirmation of work done by Gill and reviewed above. We have thus shown the individual reaction of each of the salts as related to the nitrate determination and the causes which are responsible for the difference. Nitric acid is evidently set free from nitrates through the combined action of heat and the  $\text{SO}_4$  radicle on the steam bath and in the evolution of chlorine when the phenoldisulphonic acid is added to nitrate and chloride-containing material.  $\text{Na}_2\text{CO}_3$ , however, possessing only a weak and unstable acid radicle is powerless to set free nitric acid either through the help of heat on the steam bath or by its reaction with the phenoldisulphonic acid.

#### GENERAL REMARKS

So many factors may interfere with the determination of nitrates by the phenoldisulphonic acid method that it would appear to be almost worthless, and yet it would seem to us that since there is no other good method to take its place which is nearly as simple and capable of use in very numerous determinations, it is worth while taking certain precautions to avoid

error, and to establish the method on a firmer basis. Our results as above outlined show that losses of nitrates are induced by the presence of  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$ , and such losses are indeed hard to avoid when working with "alkali soils." Even the suggestion of Chamot that  $\text{AgSO}_4$  might be used to precipitate chlorides would seem, from our results, not to be useful, since the addition of sulfate to the solution would accomplish very considerable losses itself, even if the silver sulfate can be obtained nitrate-free, which Chamot claims is seldom the case. So that while we deem it unsafe in the presence of considerable quantities of salts containing chlorides and sulfates to determine nitrates by the phenoldisulphonic acid method and would therefore recommend the Street modification of the Ulsch method in such cases, it is likewise clear that many of the nitrate determinations made in soil laboratories, as is especially the case in soil bacteriological work, would not be interfered with by salts. In such cases the method can be safely depended on if potash alum, aluminum cream, and bone black are not used to coagulate clay and organic matter, since they have been found in the researches above described to be productive of very serious errors. We recommend as a substitute for these coagulating agents the oxide of lime in its chemically pure state, to be employed in accordance with the method above given. The losses of nitrates sustained through its use have been shown to be very small in the work above reported, and it may be employed by grinding the soil with water or by direct addition to the muddy suspension prepared from the soil.

Other sources of loss such as those brought about through the sterilization of controls in the autoclave are unavoidable. They have been found at times to be distinctly appreciable, and especially in the presence of considerable quantities of organic matter. It is further of the greatest interest to learn, from the experiments above described, of the action of the anion of the salts employed in our studies and the losses of nitrates occurring on the water bath from solutions being evaporated there when either  $\text{NaCl}$  or  $\text{Na}_2\text{SO}_4$  is present.

We should also make mention here of our attitude toward the use of  $\text{NH}_4\text{OH}$  instead of  $\text{KOH}$ , which was found superior



to the former in the investigations above reviewed. While higher absolute results may no doubt be obtained from the use of KOH than from  $\text{NH}_4\text{OH}$ , and while in addition ammonia possesses other objectionable features, we were not aware of the first of these objections when these investigations were begun and did not deem the others serious enough to warrant a change in the established method. Moreover, the same relative values would exist for the data above given if obtained with one or the other of the hydrates, and therefore our results, having been obtained throughout by the use of ammonia, do not in any way lose their value. We do intend, however, in the future to employ KOH exclusively in nitrate determinations made in this laboratory. Finally we desire to call the attention of soil chemists to the fact that losses of nitrates by the agencies above described never seem to occur in accordance with any definite law, with the exception of the case in which the various alkali chlorides are compared. In these it would appear, from calculations which we have made, that the losses of nitrates are proportional to the amounts of chlorine present. While no law can be formulated, however, in accordance with which nitrates are lost in the presence of salts, it may be possible to work out tables for the losses of nitrates incurred in the presence of varying quantities of chlorides and sulphates, and to make corrections, therefore, in samples whose composition is unknown after alkali determinations are made. It is true, however, that calculation has shown on the basis of data in Table VIII that the losses of nitrates induced by chlorides alone are proportional to the amount of chlorine present.

### CONCLUSIONS

1. The "alkali" salts  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$  induce losses of nitrates when the latter are determined by the phenoldisulphonic acid method.  $\text{Na}_2\text{CO}_3$  has no such effect.  $\text{NaCl}$  induces much greater losses than  $\text{Na}_2\text{SO}_4$ .

2. Among the substances used to coagulate clay and organic matter from solutions in which nitrates are to be determined, potash alum, aluminum cream, and bone black have been found decidedly unreliable. They all induce large losses of nitrates.

3. Lime has been found to be much more reliable for the purpose named than any of the other substances, the losses incurred through its use being very small.

4. The reason for the difference between the action of  $\text{Na}_2\text{SO}_4$  and  $\text{Na}_2\text{CO}_3$  so far as the nitrate losses are concerned is to be found in the fact that  $\text{Na}_2\text{SO}_4$  induces the loss of nitric acid from the solution while the latter is being evaporated, while  $\text{Na}_2\text{CO}_3$  containing only a weak acid radicle has no power to set nitric acid free. Neither  $\text{Na}_2\text{SO}_4$  nor  $\text{Na}_2\text{CO}_3$  has the power to set nitric acid free from nitrates when the dry residues of the two are mixed prior to treatment with phenoldisulphonic acid.

5. Losses of nitrates from solutions as induced by chlorides alone seem to be proportional to the amount of chlorine present.

6. The work of Gill which showed that chlorine induces losses both on the water bath and in mixing the dry residue with phenoldisulphonic acid is confirmed.



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THE EFFECTS OF CALCIUM AND  
MAGNESIUM CARBONATES ON SOME BIO-  
LOGICAL TRANSFORMATIONS OF  
NITROGEN IN SOILS

BY  
W. P. KELLEY

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## HISTORICAL INTRODUCTION

Loew<sup>1</sup> and his co-workers found, some years ago, that the growth of a number of plants may be markedly influenced by variations in the ratio of calcium to magnesium, both in solution and soil cultures. Osterhout<sup>2</sup> also showed that a more or less definite relation between other elements in culture solutions is necessary for maximum growth. These and other researches have drawn attention to certain long neglected phases of plant physiology and strengthen the view that in addition to the mere presence of the necessary elements, plants also demand a physiologically balanced relation between the elements in solution if maximum growth is to be produced. By means of artificial culture solutions principles of great importance are being worked out, but in generalizing from culture solutions to natural soils, many difficulties arise. The great complexity of the factors involved and the difficulties inherent in the question necessitate the greatest care in making broad generalizations regarding soils.

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<sup>1</sup> Loew and May, Bur. Plant Ind. U. S. D. A., Bul. No. 1; Aso, Bul. Col. Agr. Tokyo, vol. 4, pp. 361-370; vol. 5, p. 495; vol. 6, p. 97; Loew and Aso, vol. 7, pp. 395-407.

<sup>2</sup> Bot. Gaz. 42, 127-134; 44, 259-272; 48, 98-104.



In this connection the "lime-magnesia ratio" has become a matter of general interest and is being extensively investigated at the present time. Some recent experiments by Lemmermann<sup>3</sup> and others seem to indicate that a wide variation in this ratio is of no consequence to plants. It is well known, however, that the effects produced by natural limes and limestones are not always equal. In certain instances dolomitic limes are known to produce less favorable results than non-magnesian limes. During recent years additional light on the action of lime in soils has been found in the fact that calcium carbonate enhances certain biological activities through supplying an active base by means of which the essential neutral condition is maintained. In this connection the question of the effects on bacterial activity brought about by different sources of lime and limestone naturally suggests itself.

In regard to physiologically balanced solutions for bacteria, Dr. C. B. Lipman<sup>4</sup> has shown that the ammonification of peptone by pure cultures of *B. subtilis* is favored on the one hand by a certain ratio of calcium to potassium, magnesium to sodium and potassium to sodium; while on the other hand, he failed to observe any antagonism between calcium and magnesium or calcium and sodium. In his investigations Lipman found that a certain concentration of magnesium chloride proved toxic to the development and activity of *B. subtilis* and at the same time the addition of certain amounts of calcium chloride failed to overcome this toxicity. Likewise, magnesium or sodium was ineffective in overcoming the toxicity of calcium. While it is probably true that calcium is not necessary for the normal development of bacteria, the importance of these observations, if found to apply in soils, is at once obvious.

From a study of the effects of various carbonates on the nitrification of ammonium sulphate in solutions, Owen<sup>5</sup> in 1908 concluded that magnesium carbonate is better suited to the stimulation and growth of nitrifying organisms than calcium, potas-

<sup>3</sup> Landw. Jahrb., 40 (1911), pp. 173-254; Also see Gile, Porto Rico Sta. Ann. Rept., 1911.

<sup>4</sup> Bot. Gaz., 48 (1908), pp. 105-125; 49 (1909), pp. 41-50.

<sup>5</sup> Ga. Sta. Bul. 81 (Technical Series No. 1), 1908.

sium or ammonium carbonates. It is but fair to mention in this connection, however, that great dilution of these carbonates was employed.

In 1910 Dr. J. G. Lipman<sup>6</sup> observed that the addition of one gram of calcium carbonate per one hundred grams of a New Jersey soil stimulated the ammonification of dried blood but depressed the formation of ammonia from cotton seed meal. In parallel experiments he observed that an equal amount of magnesium carbonate caused a depression in the ammonification of dried blood but stimulated the ammonification of cotton seed meal. In other words, the ammonification of dried blood and cotton seed meal in one and the same soil were affected by calcium and magnesium carbonates in opposite ways, both as regards the carbonates and the nitrogenous substances employed. These results are interesting and suggestive and point to the complexity of this single step in the preparation of available nitrogen from the organic substances occurring in soils.

In the same year Kellerman and Robinson<sup>7</sup> pointed out that the addition of magnesium carbonate to a highly magnesian soil in quantities above 0.25 per cent greatly depressed the formation of nitrates while the application of calcium carbonate in quantities up to 2 per cent markedly stimulated nitrification. The growth of crops on this soil had been found to be much more favorably influenced by the application of ground oyster shells than by magnesium limestone. The authors inferred from their experiments that the inferior effects on crops following the application of dolomitic limestone may be due, in part, to retarded nitrification.

## AMMONIFICATION

In the course of some studies on soil bacteriology at the University of California, the writer undertook a study of certain biological transformations, as affected in two different sandy soils from California by varying amounts and combinations of calcium and magnesium carbonates. On account of the striking nature of the results obtained in the preliminary ammonification ex-

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<sup>6</sup> N. J. Sta. Rept., 1910.

<sup>7</sup> Science, 32, p. 159.

periments a systematic study of this question was undertaken. The soil employed in the ammonification experiments presently to be described was of a light sandy character having been taken from near Oakley in the upper part of San Joaquin Valley and represents a large area now devoted to the growth of peaches and other fruits. With suitable moisture conditions this land produces excellent growth of the crops suited to it. The following analysis furnished by the courtesy of Dr. Lipman sets forth the composition of this soil.

TABLE I. COMPOSITION OF SOIL USED IN AMMONIFICATION EXPERIMENTS

	Per cent
Insoluble matter .....	80.45
Soluble silica .....	6.15
Potash ( $K_2O$ ) .....	0.35
Soda ( $Na_2O$ ) .....	0.15
Lime ( $CaO$ ) .....	1.41
Magnesia ( $MgO$ ) .....	0.33
Br. Ox. Manganese ( $Mn_2O_3$ ) .....	0.09
Ferric Oxide ( $Fe_2O_3$ ) .....	3.96
Alumina ( $Al_2O_3$ ) .....	4.45
Phosphoric Acid ( $P_2O_5$ ) .....	0.10
Sulphuric Acid ( $SO_3$ ) .....	0.06
$H_2O$ at $110^\circ C.$ .....	0.80
Volatile matter .....	2.02
Total .....	100.32

In the ammonification experiments dried blood was used as a source of nitrogen. Five grams of this material and varying amounts of calcium and magnesium carbonates were thoroughly mixed with 100 gram portions of sifted soil, placed in tumblers and then optimum moisture conditions provided by the addition of sterile water. The tumblers were covered with Petri dishes and after an incubation period of seven days the ammonia was distilled into standard acid by the use of magnesium oxide and measured in the usual way. The results are recorded in the following table.

TABLE II. EFFECTS OF CALCIUM AND MAGNESIUM CARBONATES ON THE AMMONIFICATION OF DRIED BLOOD

Treatment	Ammonia nitrogen mgs.
None .....	81.4
1 Gram Calcium Carbonate .....	84.3
2 Gram Calcium Carbonate .....	85.0
4 Gram Calcium Carbonate .....	91.0
6 Gram Calcium Carbonate .....	91.0
8 Gram Calcium Carbonate .....	87.8
12 Gram Calcium Carbonate .....	87.8
1 Gram Magnesium Carbonate .....	53.2
2 Gram Magnesium Carbonate .....	53.9
4 Gram Magnesium Carbonate .....	50.0

These data, as all others submitted in this paper, represent averages of closely agreeing duplicates. In examining the above data we note a slight stimulation in ammonia formation from the use of the several amounts of calcium carbonate employed, the maximum stimulation being reached with from 4 to 6 grams per 100 grams of soil. With the use of magnesium carbonate a marked depression in ammonia accumulation occurred, there having been found to be a falling off of approximately one-third as compared with the amounts found without the use of carbonate. It is also noteworthy that one gram of magnesium carbonate proved to be about as toxic to ammonification as larger amounts.

A second series was prepared with the use of still smaller amounts of magnesium carbonate for the purpose of determining the concentration at which toxic effects begin and also to determine the minimum amount of this carbonate necessary to produce maximum toxicity. The results follow.

TABLE III. AMMONIFICATION OF DRIED BLOOD AS AFFECTED BY SMALL AMOUNTS OF MAGNESIUM CARBONATE

Treatment	Ammonia nitrogen mgs.
None .....	93.1
0.1 Gram Magnesium Carbonate .....	77.4
0.2 Gram Magnesium Carbonate .....	70.6
0.4 Gram Magnesium Carbonate .....	65.6
0.6 Gram Magnesium Carbonate .....	65.2
0.8 Gram Magnesium Carbonate .....	64.6
1.0 Gram Magnesium Carbonate .....	62.0

These data are instructive as showing the marked depression of ammonification in the soil employed, even with the small amount of .1 per cent of magnesium carbonate. The toxicity increased with greater amounts of the magnesium carbonate added reaching a practical maximum with from 0.8 to 1 gram per 100 grams of soil.

According to Loew the toxic effects of an excess of magnesia in soils can be overcome or antagonized by the application of lime. While this theory was proposed and held for the higher plants, it was thought to be of some interest to study the question with reference to the ammonification process. Accordingly the following series of experiments was arranged. In these trials one gram of magnesium carbonate per 100 grams of soil was used throughout, this quantity having been found to be the lowest that produced maximum toxicity.

TABLE IV. AMMONIFICATION OF DRIED BLOOD IN THE PRESENCE OF BOTH  $\text{CaCO}_3$  AND  $\text{MgCO}_3$

Treatment	Ammonia nitrogen mgs.
1 Gram Calcium Carbonate .....	84.3
1 Gram Magnesium Carbonate .....	53.9
1 Gram Magnesium Carbonate + 0.5 Grams Calcium Carbonate	51.1
1 Gram Magnesium Carbonate + 1. Grams Calcium Carbonate	53.9
1 Gram Magnesium Carbonate + 2. Grams Calcium Carbonate	53.2
1 Gram Magnesium Carbonate + 3. Grams Calcium Carbonate	50.6
1 Gram Magnesium Carbonate + 4. Grams Calcium Carbonate	51.1
1 Gram Magnesium Carbonate + 5. Grams Calcium Carbonate	50.7
1 Gram Magnesium Carbonate + 6. Grams Calcium Carbonate	50.3
1 Gram Magnesium Carbonate + 8. Grams Calcium Carbonate	50.7
1 Gram Magnesium Carbonate + 12. Grams Calcium Carbonate	50.4

From these data it is at once seen that no antagonism was produced. Even the very large amount of 12 grams of calcium carbonate in no way reduced the toxic effects produced by one gram of magnesium carbonate. The results, therefore, are in harmony with the observations made by Dr. C. B. Lipman<sup>8</sup> in his studies on the physiology of *B. subtilis*.

<sup>8</sup> *Loc. cit.*

## NITRIFICATION

Having failed to observe any antagonism between calcium and magnesium in the complex process of ammonification in the soil under investigation, attention was directed to a study of nitrification under similar conditions. A sandy soil from Anaheim, California, that contained a vigorous nitrifying flora, was employed in these studies. The following table of analyses furnished by the kindness of Dr. Lipman shows the chemical composition of this soil.

TABLE V. COMPOSITION OF SOIL USED IN NITRIFICATION EXPERIMENTS

	Per cent
Insoluble matter .....	73.59
Soluble Silica .....	11.17
Potash ( $K_2O$ ) .....	.64
Soda ( $Na_2O$ ) .....	.15
Lime ( $CaO$ ) .....	1.39
Magnesia ( $MgO$ ) .....	.93
Br. Ox. Manganese ( $Mn_2O_3$ ) ..	.04
Ferric Oxide ( $Fe_2O_3$ ) .....	5.10
Alumina ( $Al_2O_3$ ) .....	3.92
Phosphoric Acid ( $P_2O_5$ ) .....	.12
Sulphuric Acid ( $SO_3$ ) .....	.02
Volatile matter } .....	2.88
$H_2O$ at $110^\circ C$ }	
Total .....	99.95

The nitrification experiments were carried out in tumblers, two grams of dried blood being mixed with each 100 gram portion of soil. The amounts of calcium and magnesium carbonates added are shown in the table. Optimum moisture conditions were maintained throughout the 21 day incubation period during which time a temperature of 27 to 28 degrees was maintained. The results are shown in the following table.

TABLE VI. EFFECTS OF CALCIUM AND MAGNESIUM CARBONATES ON THE NITRIFICATION OF DRIED BLOOD

Treatment	Nitrate nitrogen found mgs.
None .....	14.5
1.0 Gram Calcium Carbonate .....	23.5
2.0 Gram Calcium Carbonate .....	19.2
4.0 Gram Calcium Carbonate .....	21.2
8.0 Gram Calcium Carbonate .....	20.2
0.1 Gram Magnesium Carbonate .....	3.6
0.2 Gram Magnesium Carbonate .....	2.9
0.4 Gram Magnesium Carbonate .....	2.8
0.8 Gram Magnesium Carbonate .....	5.1
1.0 Gram Magnesium Carbonate .....	1.0
2.0 Gram Magnesium Carbonate .....	2.0
4.0 Gram Magnesium Carbonate .....	2.9
8.0 Gram Magnesium Carbonate .....	3.3
Original soil .....	5.0

It will be observed that while approximately a 50 per cent stimulation in nitrate formation was effected by the addition of calcium carbonate, nitrification was totally inhibited by the addition of one-tenth of one gram of magnesium carbonate. Before further discussing these results the data obtained from the effects of calcium and magnesium carbonates acting synchronously will be presented.

TABLE VII. THE LACK OF ANTAGONISM BETWEEN CALCIUM AND MAGNESIUM CARBONATES AS SHOWN IN THE NITRIFICATION OF DRIED BLOOD

Treatment	Nitrate nitrogen found mgs.
None .....	14.5
1.0 gram Calcium Carbonate .....	23.5
0.1 gram Magnesium Carbonate .....	3.6
0.1 gram Magnesium Carbonate and 1. gram Calcium Carbonate .....	4.1
0.1 gram Magnesium Carbonate and 2. gram Calcium Carbonate .....	3.4
0.1 gram Magnesium Carbonate and 3. gram Calcium Carbonate .....	2.6
0.2 gram Magnesium Carbonate and 1. gram Calcium Carbonate .....	1.9
0.2 gram Magnesium Carbonate and 2. gram Calcium Carbonate .....	1.4
0.2 gram Magnesium Carbonate and 3. gram Calcium Carbonate .....	2.0
0.4 gram Magnesium Carbonate and 1. gram Calcium Carbonate .....	2.2
0.4 gram Magnesium Carbonate and 2. gram Calcium Carbonate .....	1.8
0.4 gram Magnesium Carbonate and 3. gram Calcium Carbonate .....	3.1
0.8 gram Magnesium Carbonate and 1. gram Calcium Carbonate .....	2.9
0.8 gram Magnesium Carbonate and 2. gram Calcium Carbonate .....	3.5
0.8 gram Magnesium Carbonate and 3. gram Calcium Carbonate .....	4.1

Here again it is shown that .1 gram of magnesium carbonate per 100 grams of soil entirely prevented nitrification. Neither do we observe any effective antagonism through the use of calcium carbonate.

On the one hand it was found that ammonification of dried blood was seriously interfered with by the presence of small amounts of magnesium carbonate, and on the other, nitrification was completely prevented by its presence. In neither case was there any evidence of an antagonism between magnesium and calcium carbonates. In the above nitrification experiments, magnesium carbonate not only prevented the formation of nitrates but at the same time induced a reduction in the amounts of nitrates originally present in the soil. It was observed that with the addition of magnesium carbonate a much more abundant growth of moulds took place than in the tumblers receiving calcium carbonate.

With a view of throwing further light on this question, total nitrogen was determined, both before and after the incubation period of 21 days, in a similar set of experiments to which one gram of magnesium carbonate had been added. The result showed that during the period of bacterial action, similar to that in the preceeding nitrification experiments, the soil sustained a loss equal to about 20 per cent of the combined nitrogen originally present.

Two factors suggest themselves as bearing on this question. The first and probably most important is that of volatilization and, therefore, loss of ammonia. J. G. Lipman<sup>o</sup> in his numerous researches found that the dilution of a heavy silt loam with silica sand caused a loss of ammonia in ammonification experiments. The loss began to manifest itself with the use of 30 per cent of sand but greatly increased with larger amounts. This loss was attributed to the volatilization of ammonia and was sufficiently great to give an appreciable odor of ammonia above the tumblers. The soils employed in the experiments herein described were largely composed of sand and contained very small amounts of silt and clay. The substances capable of fixing large amounts of ammonia are, therefore, largely absent from

<sup>o</sup> N. J. Sta. Rept., 1909.



these soils and consequently considerable loss may have been sustained through the volatilization of ammonia. The data on ammonification, therefore, should be considered as representing the ammonia accumulated rather than the absolute amounts formed. The relative effects of calcium and magnesium carbonates on the loss of ammonia were not investigated.

A second factor in the loss of nitrogen is that of denitrification. It was recently shown by Vogel<sup>10</sup> that calcium carbonate under certain conditions can bring about a considerable loss of nitrogen as nitrates in soils through denitrification. In the previous experiments it was observed that with the use of magnesium carbonate a decided reaction for nitrites could be obtained. Denitrification, therefore, took place and an actual loss of nitrogen is probably traceable to this cause. From the preceding data (Table VI) it is seen that the use of small amounts of magnesium carbonate not only inhibited nitrification but, as previously mentioned, also caused a considerable loss of the nitrates already in the soil. We have here, therefore, still further evidence of denitrification having taken place. With the use of larger amounts of magnesium carbonate, nitrification and denitrification were both inhibited but no considerable loss of the nitrates originally present in the soil took place.

It seems probable, therefore, that the smaller amounts of magnesium carbonate were toxic to the nitrifying bacteria while still allowing the denitrifiers to act, but under the influence of larger amounts of magnesium carbonate both the nitrifying and denitrifying groups were rendered inactive.

The striking nature of the results obtained in the previous ammonification and nitrification experiments suggested a study of nitrogen fixation under similar conditions. For this purpose the Anaheim soil was employed since it contains a vigorous nitrogen fixing flora. Mannite was used in these experiments and the usual method followed. The results obtained proved to be so irregular and discordant that their publication is withheld at this time. In one series a slight decrease in the amount of nitrogen fixed followed the use of magnesium carbonate, while in still another series no effects were observed.

<sup>10</sup> Centbl. Bakt. 34, pp. 540-561.

## DISCUSSION

The experimental data presented above show that under the conditions employed and in the soils studied, calcium carbonate stimulated the ammonification of dried blood to a limited extent but exercised a more noteworthy stimulating effect on nitrification. With magnesium carbonate a pronounced toxic effect was produced. In the ammonification of dried blood there was sustained a loss of about one-third as compared with the experiments without the use of carbonates, while in the nitrification experiments magnesium carbonate completely inhibited nitrate formation. It is also noteworthy that no evidence of antagonism between calcium and magnesium carbonates was observed. It is not intended, however, to generalize from these results. It does not follow that similar results would be obtained from any soil. In fact, data already obtained from other soils show that the phenomena observed in the two soils above discussed are not of universal occurrence under similar conditions.

A further study of the lime-magnesia ratio in reference to nitrogen transformations in soils is now under way with the use of several types of Hawaiian soils and interesting results have already been obtained. A more complete interpretation of the results obtained is reserved for a subsequent publication after a wider range of observations have been made. Before a satisfactory understanding of the lime-magnesia question in regard to field crops is presented it is imperative that we have more specific knowledge concerning the effects produced on the various organisms of soils, now generally admitted to be of fundamental importance in plant growth, and it is especially important that the effects produced on the organisms affecting nitrogen transformations be more fully understood. It is hoped that this work may stimulate other investigations along this line.

The author wishes to extend his thanks to Dr. C. B. Lipman, in whose laboratory this work was carried out, for many valuable suggestions offered from time to time and the great interest shown.



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**THE ALUMINUM REDUCTION METHOD AS  
APPLIED TO THE DETERMINATION OF  
NITRATES IN "ALKALI" SOILS**

**BY**  
**PAUL S. BURGESS**

**UNIVERSITY OF CALIFORNIA PRESS**  
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THE ALUMINUM REDUCTION METHOD AS  
APPLIED TO THE DETERMINATION OF  
NITRATES IN "ALKALI" SOILS

BY  
PAUL S. BURGESS

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While the phenoldisulphonic acid method for determining nitrates in soils at present offers the most speedy and satisfactory means of ascertaining the nitrate content of soils free from "alkali," it has been shown<sup>1</sup> to be of questionable value when employed with soils containing even small amounts of soluble salts, and especially in the presence of the chlorides and sulfates of the alkalies. Since this is true, and further since the number of nitrate determinations on soils containing "alkali" is constantly increasing, due to the great increase in our research work on "alkali" problems, both chemical and bacteriological, it was deemed by the writer to be a matter of importance to establish a method for the determination of nitrates which was not affected by the presence of soluble salts.

The possible methods considered were Busch's "nitron" process, the methods depending upon the liberation and subsequent measuring of nitric oxide, and the reduction methods in which the nitrate nitrogen is reduced to ammonia and either titrated against a standard acid solution or Nesslerized.

Busch's "nitron" process<sup>2</sup> was rejected for the following reasons. A number of the acids (both organic and inorganic),

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<sup>1</sup> Univ. of Calif. Publ. Agr. Sci., vol. 1, no. 2, pp. 21-37. Utah Agr. Exp. Sta. Bull., 106.

<sup>2</sup> Ber. Deut. Chem. Gesell. 38 (1905), 3, pp. 861-866.



and salts found in soils, form insoluble compounds with the "nitron" (1.4-diphenyl-3.5 endanilodihydrotryazol) as well as does the nitrate radicle. The sterilization of soils also appears to liberate substances which interfere with the crystallization of the "nitron" nitrate.<sup>3</sup> The occurrence of soluble organic matter invariably present in the soil solutions tends, in many cases, to vitiate completely the results.<sup>4</sup> The calcium oxid, which we use to coagulate the clay before filtration, comes down as the carbonate in the filtrate, thus making a gravimetric determination impracticable. (Lime has been shown to effect the *least* loss of nitrates of any of the common coagulants.) The trouble and cost of procuring the reagent also militated against the use of this method. The Schulze-Tiemann,<sup>5</sup> Schlösing-Wagner,<sup>6</sup> and similar methods which depend upon the liberation and subsequent measurement of the nitric oxide from the nitrates did not appear feasible because of the errors introduced through atmospheric conditions, the expense of apparatus for a large number of determinations, and the length of time necessary for the operations involved.

Thus all but the reduction methods were eliminated. In 1890 the Agricultural Experiment Station<sup>7</sup> at Halle, Germany, perfected a reduction method for the determination of nitrogen in nitrates. They used zinc dust, iron filings, and a solution (sp. gr. 1.3) of sodium hydrate. The presence of chlorides and sulfates did not impair the accuracy of the determination. Several modifications of this reduction process are now used. In the modified Ulsch<sup>8</sup> method sulfuric acid and reduced iron are employed to liberate the nascent hydrogen, an excess of magnesium oxid being added just before distillation. In the Devarda<sup>9</sup> method an alkali, an alloy of aluminum, copper and zinc, and ethyl alcohol are all used to effect the reduction. M. E. Pozzi-

<sup>3</sup> J. Litzendorff, *Ztschr. Angew. Chem.* 20 (1907), 51, pp. 2209-13.

<sup>4</sup> *Mich. Exp. Sta. Rep. for 1911*, pp. 178-181.

<sup>5</sup> Bohm, *Ztschr. Zuckerind.* 25 (1900), p. 356, abs. in *Chem. Centrbl.* (1901), I, 22, p. 1216.

<sup>6</sup> König's *Untersuch. Landw. Stoffe*, p. 151.

<sup>7</sup> *Experiment Station Record*, vol. V, pp. 464-465.

<sup>8</sup> *New Jersey Exp. Sta. Rep. for 1892*, pp. 188-193.

<sup>9</sup> *Analyst*, 35 (1910), 412, p. 307.

Escot<sup>10</sup> utilizes aluminum filings, mercuric chloride and a solution of potassium hydrate. J. T. Bornwater<sup>11</sup> uses aluminum filings and a solution of potassium hydrate.

Still further elimination was thus necessary among the reduction methods. The modified Ulsch method presented certain difficulties of technique which, in the case of numerous determinations, would render the method impracticable. The aluminum reduction method has been heretofore used successfully in water analysis,<sup>12</sup> and it appeared advisable to make a study of it as applied to determinations of large amounts of nitrates as met with in soil work; it further seemed important to determine the feasibility of its use for a large number of determinations made simultaneously. Such other factors also as the amounts of aluminum and of alkali to employ, the length of time, and the temperature for reduction, demanded a careful test.

#### DESCRIPTION OF THE METHOD

One hundred grams of the soil in which the nitrates are to be determined are placed in round-bottomed, enameled cereal dishes which have a capacity of about 800 c.c. Mortars were used at first, but the cereal dishes were found to be much lighter, easier to manipulate, and less expensive. Two grams of powdered CaO and exactly 200 c.c. of distilled water are added to each dish. The contents of each dish are now thoroughly ground and mixed with a pestle for from 3 to 5 minutes, after which the soil and clay are allowed to settle for 15 or 20 minutes, and are then filtered through paper. It may be said here that the solutions should never be allowed to stand over 2 to 3 hours, as there occurs a noticeable loss of nitrates, possibly due to dentrification. In case it is impossible to proceed at once with the determination a few drops of chloroform<sup>13</sup> may be added. One hundred c.c. portions of the filtrates, obtained as above described, are placed in 400 c.c. casseroles and 2 c.c. of a 50 per cent NaOH solution, free from nitrates, added to each. These are then boiled

<sup>10</sup> Ann. (Chim. Analyt. 14 (1909), 12, pp. 445-446.

<sup>11</sup> Chem. Centbl. (1906), I, 8, p. 703.

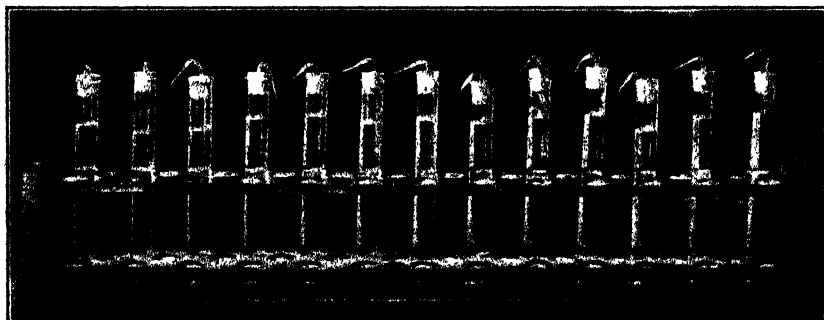
<sup>12</sup> Amer. Jour. Pub. Hyg., vol. XIX, 3, p. 1.

<sup>13</sup> V. I. Sazanov, Abs. in Centbl. Zuckerindus. 15 (1907), 34, p. 923.

down to about half their original volume to drive off ammonia, the residues washed into 125 c.c. Jena test tubes, diluted to 100 c.c., and a strip of aluminum (about  $150 \times 6 \times .4$  mm., weighing approximately one gram) added to each. The tubes are then stoppered with one-hole rubber stoppers carrying bent glass tubes, each of which has been drawn out to a fine capillary tip. The solutions are allowed to remain in the tubes from 11 to 14 hours at a constant temperature of from  $20^{\circ}$  to  $22^{\circ}$  C. (about our laboratory temperature). In case of large amounts of nitrogen the temperature is of prime importance, a lower temperature giving incomplete reduction in the above mentioned time, while a higher temperature may induce a considerable loss of ammonia. After reduction the contents of the test tubes are washed into distilling flasks, about 300 c.c. of distilled, ammonia-free water is added, and the ammonia distilled off and caught in N/10 HCl, the excess of acid being titrated against N/10  $\text{NH}_4\text{OH}$ .

We run 24 or 36 determinations simultaneously, avoiding a loss of time by allowing the reduction to take place over night, either in an incubator kept at  $20^{\circ}$ - $22^{\circ}$  C., or when temperature conditions are right, in the laboratory.

The cut below shown is a photograph of a rack which the writer had made for the purpose of holding 36 of the large test tubes while reduction was taking place. The holes are all numbered, thus doing away with the necessity of marking the glass tubes, the determinations being run in rotation.



## TEST OF THE REDUCTION METHOD WITH LARGE QUANTITIES OF NITRATE

In soil work considerable amounts of nitrates are often encountered. The first series of experiments were thus made to determine whether or not the reduction method would prove accurate in the presence of from 30 to 60 mgs. of nitrate nitrogen.

The results set forth in the following table are averages of several analyses made at the same time and under similar conditions. A comparison is also made here of the reduction method with the phenoldisulphonic acid method.

### TABLE I

A COMPARISON OF THE REDUCTION METHOD AND THE PHENOLDISULPHONIC  
ACID METHOD WITH LARGE AMOUNTS OF NITRATE NITROGEN

No.	Nitrate N added Mgs.	Reduction Method Nitrate N recovered Mgs.	Phenoldisulphonic Acid Method Nitrate N recovered Mgs.
1	100	97.86	97.00
2	50	49.00	48.25
3	25	24.22	25.40

We thus see that the reduction method, even where *no* salts are present, but where large amounts of nitrates are found, is slightly more accurate than the phenoldisulphonic acid method.

### THE EFFECT OF "ALKALI" SALTS ON THE REDUCTION METHOD METHOD

As stated above, in our research work on different "alkali" problems in soils, more especially in soil bacteriology, and plant physiology, considerable amounts of salts are often used in soils in which later the nitrate content must be ascertained. Besides it is frequently necessary to determine nitrates in soils of the arid regions which contain considerable quantities of "alkali." It is important therefore to ascertain if the method herein proposed is in any wise affected by salts. Therefore the following tests were carried out. The salts employed were "Baker's An-

alyzed Chemicals." All the reagents upon analysis were found to be free from nitrogen. The salts were added in solution from accurately graduated pipettes and burettes. The reduction period for all the samples was 11 hours at a temperature of 20°-22° C.

The following table shows the results obtained in a series of nitrate determinations made by the reduction method in the presence of "alkali." The details with reference to these determinations are also given in the table, and the figures are averages of closely agreeing duplicate or triplicate analyses. This statement applies also to the following tables.

TABLE II

## EFFECTS OF NaCl

NaCl added Grams	Nitrate N added Mgs.	Aluminum Reduction Method Nitrate N recovered Mgs.	Phenoldisulphonic Acid Method Nitrate N recovered Mgs.
0.2	100	97.90	79.50
0.2	50	49.10	43.00
0.2	25	24.50	20.75

EFFECTS OF Na<sub>2</sub>SO<sub>4</sub>

Na <sub>2</sub> SO <sub>4</sub> added Grams	Nitrate N added Mgs.	Aluminum Reduction Method Nitrate N recovered Mgs.	Phenoldisulphonic Acid Method Nitrate N recovered Mgs.
0.35	100	98.42	72.75
0.35	50	49.42	38.20
0.35	25	24.36	19.75

EFFECTS OF Na<sub>2</sub>CO<sub>3</sub>

Na <sub>2</sub> CO <sub>3</sub> added Grams	Nitrate N added Mgs.	Aluminum Reduction Method Nitrate N recovered Mgs.	Phenoldisulphonic Acid Method Nitrate N recovered Mgs.
0.1	100	98.14	86.25
0.1	50	49.42	42.50
0.1	25	24.50	25.20

TABLE II—(Continued)  
EFFECTS OF "MIXED ALKALI" SALTS

Salts added Grams	Nitrate N added Mgs.	Reduction Method Nitrate N recovered Mgs.	Acid Method Nitrate N recovered Mgs. Phenoldisulphonic
0.2 NaCl } 0.35 Na <sub>2</sub> SO <sub>4</sub> } 0.1 Na <sub>2</sub> CO <sub>3</sub> }	100	99.54	65.00
0.2 NaCl } 0.35 Na <sub>2</sub> SO <sub>4</sub> } 0.1 Na <sub>2</sub> CO <sub>3</sub> }	50	50.05	33.00
0.2 NaCl } 0.35 Na <sub>2</sub> SO <sub>4</sub> } 0.1 Na <sub>2</sub> CO <sub>3</sub> }	25	24.57	12.00

That the salts present have little effect on the accuracy of the determination by the reduction method, and the superiority of the latter over the phenoldisulphonic acid method, is clearly shown by a comparison of the last two columns in the foregoing table. Where large losses of nitrates are induced by the presence of NaCl and Na<sub>2</sub>SO<sub>4</sub> with the second named method, there are only slight losses when the reduction method is employed, and these are found by a comparison with Table I to be apparently due to other causes.

In most cases where the reduction method is used there are slight losses of ammonia, although in a few individual analyses all of the nitrogen was recovered. It may be stated here that several analyses were run on solutions containing 100 mgs. of nitrate nitrogen, placing rubber stoppers in the large reducing test tubes, carrying bent glass tubes as traps and connecting with test tubes containing 10 c.c. of N/10 HCl each. These were later titrated against N/10 NH<sub>4</sub>OH and the results added to the figures found by distillation. By using this extra process we were able to recover all the N as NH<sub>3</sub>, but it was not found to be necessary where the smaller amounts of nitrate were present. It should be remembered that in soil work over 30 mgs. of nitrate nitrogen per 100 grams of soil are infrequently found, and of this amount only an aliquot (one-half) is taken for the actual analysis. In nitrification work also, especially where the production of nitrates is intense, the difference between parallels is

often from one to one and one-half milligrams of nitrogen, or about the amounts which appear to be lost where large quantities of nitrates are determined by this method.

As a means of comparison, the same amounts of "alkali" salts and nitrate were added and the entire series analyzed by the phenoldisulphonic acid method. The results of this work are shown in the last column of Table II. The standard method here calls for the use of 2 c.c. of the phenoldisulphonic acid, but we found that even where *no* foreign salts were present, if the amount of nitrate nitrogen exceeded about 10 mgs. 4 c.c. were necessary to complete the reaction.

The results of tests of the colorimetric method where large amounts of nitrates were present, using respectively 2 and 4 c.c. of the phenoldisulphonic acid, follow:

TABLE III

No.	"Alkali" salts present	Nitrate N added Mgs.	Nitrate N recovered using 2 c.c. phenoldisul- phonic acid Mgs.	Nitrate N recovered using 4 c.c. phenoldisul- phonic acid Mgs.
1	0	100	58.00	97.00
2	0	50	35.00	48.25
3	0	25	23.60	25.40

To ascertain whether or not 4 c.c. portions of the acid were sufficient, 6 c.c. quantities were tried. No gains in the amount of nitrate nitrogen recovered here resulted. Four c.c. portions were employed in the experiments reported in Table II.

It is interesting to note that while my results confirm the work of Lipman and Sharp on the effects of  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$  on the phenoldisulphonic acid method, they are partly at variance with them on the effects of  $\text{Na}_2\text{CO}_3$ . The investigators named found that the nitrate determination by the method mentioned was in no wise affected by  $\text{Na}_2\text{CO}_3$ , but it should be recalled that they employed comparatively small quantities of nitrates and carbonates and that in the presence of larger quantities the chances of error are magnified.

# THE EFFECT OF SOLUBLE ORGANIC MATERIALS ON THE REDUCTION METHOD

After having found that the soluble mineral salts had no effect on the accuracy of the reduction method, it was deemed advisable to ascertain whether or not the organic matter, which is always present in the soils to a greater or less extent, interferes with it. Such organic materials may be roughly divided into two great classes: the humates, or the salts of humic acid with the alkalies of the soil, and the soluble carbohydrate material. By simply tritulating the soil sample with pure water but a very small percentage of the former class of compounds is ever extracted, while soluble carbohydrates are present in soils only in exceedingly small quantities, except in rare cases. In these tests dried, water soluble humus and dextrose were used and two sets of experiments were run, in one of which the solution to be reduced contained .2 per cent of dried humus and the other of which contained 1 per cent of dextrose. The addition of humus produced a very dark brown solution, much darker in fact than any solution that can be obtained by tritulating, even soil high in humus, with water. The analytical procedure was the same as that above given and the results are expressed in Table IV, and represent averages of closely agreeing duplicates.

TABLE IV

## EFFECT OF SOLUBLE HUMUS

No.	Soluble humus added Grs.	Nitrate N added Mgs.	Nitrate N recovered Mgs.
1	0.2	100	95.62
2	0.2	50	48.86
3	0.2	25	24.36

## EFFECT OF DEXTROSE

No.	Dextrose added Grs.	Nitrate N added Mgs.	Nitrate N recovered Mgs.
1	1.0	100	96.60
2	1.0	50	48.44
3	1.0	25	24.50



These results show a slight loss where large amounts of nitrate were present, but the same also holds true above, as an examination of the foregoing tables will indicate. Therefore the losses, which are also in part explained above, are not to be attributed to the presence of excessive amounts of soluble organic materials. Besides, the losses brought about for the reasons indicated are of no serious practical import, since most soils seldom contain more than from 15 to 20 milligrams of nitrogen per 100 grams of soil.

THE TIME REQUIRED FOR COMPLETE REDUCTION AT A  
TEMPERATURE OF 20°-22° C.

That the time required for complete reduction should be accurately ascertained at a given temperature is obvious. The results given below are averages of analyses carried on simultaneously.

TABLE V  
THE EFFECT OF TIME ON THE PROCESS OF REDUCTION AT CONSTANT  
TEMPERATURE (20°-22° C.)

Hours Nitrate N added Mgs.	1	2 ½	4	6	8	9 Nitrate N recovered Mgs.	10	11	12	15	24
100	19.04	33.88	46.20	55.02	89.74	93.10	96.53	97.90	97.44	98.42	95.20
50	19.18	22.05	31.36	36.82	46.76	47.18	49.14	49.10	48.86	49.42	49.42
25	14.35	17.71	.....	19.00	20.79	23.94	23.94	24.50	24.50	24.50	24.50

In all of these samples 0.2 per cent of NaCl was present. A study of this data shows 11 to 15 hours to be the optimum time limits for reduction at 20°-22° C. A longer time results in a slight loss of ammonia from the solutions containing the larger amounts of nitrogen, while less than 11 hours is too short to effect complete reduction.

THE EFFECT OF TEMPERATURE ON THE TIME NECESSARY FOR  
REDUCTION

Temperature is of vital importance, both in the accuracy of the determination and also on the time necessary for complete reaction. This is borne out by the following table.

TABLE VI

TIME OF REDUCTION 12 HOURS. TEMPERATURE CONSTANT AT 8° TO 10° C.

NaCl added Grams	Nitrate N added Mgs.	Nitrate N recovered Mgs.
.2	100	47.53
.2	50	28.42
.2	25	22.96

TIME OF REDUCTION 6 HOURS. TEMPERATURE CONSTANT AT 45° C.

NaCl added Grams	Nitrate N added Mgs.	Nitrate N recovered Mgs.
.2	100	80.93
.2	50	43.26
.2	25	22.12

We thus see that at a temperature of 45° C. the reduction was more nearly complete in 6 hours than it was at the end of 12 hours where the temperature was maintained at 10° C. By means of a series of experiments similar to those shown in Table IV it is readily possible to find the optimum length of time for complete reaction at any given temperature.

#### AMOUNT NaOH SOLUTION REQUIRED

By another series of experiments, in which the traps above mentioned were used and different amounts (varying from 1 to 5 c.c.) of the 50 per cent NaOH solution added, we found that 2 c.c. of the latter gave the best results. One c.c. evidently did not generate hydrogen in sufficient quantities for complete reduction (especially in the presence of the larger amounts of nitrate), while the 4 and 5 c.c. portions made the solutions so alkaline as to effect the escape of considerable amounts of ammonia.

#### GENERAL REMARKS

The results obtained in the experiments described above are considered a valuable contribution to our methods of chemical and biochemical soil analysis, because they establish, on a practical basis, a method for the determination of nitrates in soils which yields accurate results, regardless of the condition of the soil which is examined. While for rapid determinations the reduction method is not found as serviceable as the phenoldisulphonic acid method, it replaces the latter for work on "alkali" bearing soils where it alone can be safely employed.

Moreover, the writer's results are also proof of the fact, as the data in Table I indicate, that even in the *absence* of salts the reduction method is superior to the colorimetric method where large quantities of nitrates are concerned.

In the case of soils containing large amounts of organic matter the reduction method is again found superior, since it is frequently found impossible to remove the color of a soil extract from such soils without a loss of nitrates.

So far as my investigations have gone on the subject, and I have tried to take into consideration the several factors involved, no uncontrollable factor has been found to militate against the successful use of the aluminum reduction method. Great accuracy can be obtained by taking the precautions above described, and reasonable accuracy may be assured in ordinary work without the use of such extraordinary measures. The method is strongly commended to the attention of soil investigators in all phases of the work and it is to be hoped that it may serve to render simple and free from annoyance the determination of nitrates in soils under circumstances which until now have presented many difficulties.

## CONCLUSIONS

1. The aluminum reduction method for the determination of nitrates in soils yields the most accurate results of all methods now commonly in vogue.

2. "Alkali" salts do not in any way interfere with the successful operation of the method.

3. The presence of extraordinarily large amounts of soluble organic materials (soluble humus and dextrose) have little effect on the method.

4. A temperature of 20° C. for from 11 to 15 hours has been found the optimum for the reduction of large quantities of nitrates.

5. The proper amount of NaOH to be employed in the reduction was found to be 2 c.c. of a 50 per cent solution, with an aluminum strip weighing approximately one gram.

My thanks are due Prof. C. B. Lipman for helpful suggestions and critical reading of the manuscript.

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STUDIES UPON INFLUENCES AFFECTING  
THE PROTEIN CONTENT OF WHEAT

BY

G. W. SHAW

UNIVERSITY OF CALIFORNIA PRESS

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In 1905 the Merchants' Exchange of San Francisco, the State Board of Trade, the Sacramento Valley Development Association, and the more prominent millers and grain dealers of California called the attention of the Agricultural Department of the University to the fact that the milling trade found it necessary to import many hundred tons of wheat per year to maintain the quality of California flour owing to the low gluten content of the wheat grown in the state.

With the view of ascertaining the causes of such an undesirable condition, investigations were begun under an appropriation made by the Legislature at the session of 1906 (Senate Bill no. 10, entitled "An Act to provide for the improvement of cereal crops of California and appropriating money therefor"). The investigations are still under way, the bill having been re-enacted at the legislative session of 1908 and again in 1910.

The chief points for study in these investigations are: (1) To determine the effect of changes of environment upon the growth of cereals, particularly as regards the composition of the wheat kernel and with special reference to the causes of the production of a low protein content; (2) to discover or produce such wheat as will yield the largest profit per acre for the farmer, and will supply the millers with wheat of superior quality; (3) to conduct similar experiments with oats, barley, *and other cereals as may be desirable*; (4) to determine the effectiveness of various methods of culture as affecting the production of cereals.

The nature of this work is such that no permanent results can be secured from one or two seasons' operations. The element of time is an all-essential one for the solution of such problems. This would be true even though the problem was merely the development of wheats giving higher yields than those now being grown in the State, but *with the introduction of the problem of increasing the gluten content*, the element of time is even of greater importance. With this in view, the experiments were so planned that while the final results could not be expected for several years, it was possible to obtain data of importance bearing upon the entire question of cereal culture each year, material progress thus being assured.

As early as 1882 Clifford Richardson called attention to the fact that wheats from the Pacific Coast were relatively low in their protein content, and numerous analyses of California grown wheats made at the California Agricultural Experiment Station under the direction of Dr. E. W. Hilgard, in the earlier years, also showed the same condition to exist. .

That this condition exists is undoubtedly true, whatever the causes may be. To bear witness to the fact the following summary is given of analyses of white wheats grown in several different states, as compared with the analyses of 149 samples of wheats of the same class grown in California in the same years. For the sake of comparison also the average results obtained from the analysis of 49 hard winter wheats grown in Kansas the same season are included, as well as those of three soft white wheats grown in Kansas from seed originally obtained from the state of Washington.

TABLE SHOWING THE PROTEIN CONTENT OF WHITE WHEATS GROWN IN  
DIFFERENT STATES IN THE SAME SEASON

State	Number of samples	Total protein <i>Dry basis</i>
California	149	9.77
Idaho	46	10.88
Nevada	34	16.18
Montana	6	14.24
Utah	10	15.20
Washington	177	12.98
Kansas (hard winter)	49	13.25
Kansas (white wheats)*	3	10.57

\* Grown in Kansas from seed secured from the State of Washington.

In practically all of the former studies of this subject the plan has been to transfer the seed from one point to another and thus change the environmental conditions, and from the results so secured to attempt to draw general conclusions. Under such conditions, changes, not merely in environment, but also in soil, were accomplished, thus introducing too many variables. In the experiments here recorded two general methods have been followed: (a) the production of numerous varieties of wheat in the field on a small area of uniform soil and varying other condition of growth than the soil, and (b) growing wheat from the same seed under the same conditions on soils of widely different origin placed under the same cultural and climatic influences. The large and important question involved is the cause or causes of the relatively low gluten content of wheat grown under California conditions. In this connection, it is particularly desirable to know whether or not such tendency to change as exists is constant; whether it is due to some climatic influence, the time of cutting, the time of seeding, the bleaching action of the sun, the effect of early and late application of moisture, the effect of cold nights, of varying amounts of sunshine during the ripening period, or to some induced or inherent condition of the soil. The work which we have conducted has aimed at the solution of all of the questions just mentioned and consisted of several parts each of which dealt with one of the questions involved.

#### VARIATION IN THE PROTEIN CONTENT OF WHEAT

*Seasonal Variation.*—It has been observed that from season to season there is a marked variation in the protein content of wheat even though grown upon the same soil, a seeming indication that the seasonal factor is considerably greater than the soil factor in protein formation. This may be shown from a number of samples grown during the course of the experiments, some of which are given below. These wheats have been grown each year on the same soil and under the same cultural conditions at the University Farm at Davis, California.



## TOTAL PROTEIN IN DRY MATTER

	1906	1907	1908	1909	1910	1911	1912
Kubanka	9.68	9.90	14.71	14.30	14.87	10.60	14.93
Crimean	10.38	11.38	11.75	13.27	12.18	10.60	12.11
Little Club Av. of 5 plats			10.29	9.78	9.98	.....	13.00

That the quantity of available nitrogen in the soil has comparatively little bearing, if any, beyond its necessity in sufficient quantity to insure the normal growth of the plants, is indicated on a series of fertilizer plats of very uniform land, discussed further on in this paper. The seasonal effect is apparent, however, from the check plats in the same series.

*Varietal Variation.*—Variation in protein content occurs between different varieties of wheat, even though the strain be a pure one and the soil conditions under which they are grown be the same. The analyses given below are from plants of pure strain grown during the course of breeding experiments in cent-gener plats under just as nearly the same conditions as it is possible to secure in the field. The soil was uniform and the plants were 4 inches apart each way and seeded at a uniform depth of 2 inches.

## GROWN AT DAVIS, 1908

	Number of samples	Per cent protein
Little Club	163	11.83
Propo	38	12.20
Fretes	111	13.46

## GROWN AT CERES, 1908

	Number of samples	Per cent protein
Chul	53	14.98
Kharkov	50	13.95
White Australian	70	14.69

In addition to the varietal variations here shown, the above figures are of interest in showing that Kharkov, which is a hard winter wheat, when grown under California conditions in the same season does not carry a higher percent of protein than a good quality of the White Australian belonging to the white wheat class.

*Variation in Individual Plants.*—That there is a wide difference in the ability of individual plants to elaborate protein in the grain is shown from the analysis of a number of varieties grown under like conditions on the same character of soils. These plants were all grown in the same season in centgener plats, the plants being four (4) inches apart each way, and seeded at uniform depth by means of a centgener planter. The outside rows were all cut away at harvest and discarded, so that none of the plants shown here represent outside rows, but all had a uniform feeding area on uniform land.

In each of the lots shown in Table 1 it will be noted that there is a great variation between individual plants even when grown under exactly the same climatic and cultural conditions. The variation in individual plants of the variety White Australian ranged from a minimum of 9.06 to 15.31 per cent total protein, or a variation of 6.25 per cent within 25 plants, and the range in Little Club is even greater, being from 7.12 to 15.22.

Such evidence as that presented above would seem to throw the main factors determining the protein content of wheats, and inferentially other grains, externally, primarily upon some climatic influence, and secondarily upon the internal factors of variety and individuality of the plant itself. It seems certain that the individuality of the wheat plant is just as potent in determining the protein content of the grain as is the individuality of the dairy cow in determining the fat content of her milk and that it is just as impossible to feed protein into wheat by increasing the available nitrogen of the soil as it is to feed butter-fat into a cow's milk.

Following these general considerations of some of the external and internal factors bearing upon the question of protein variation, attention is turned to more restricted phases of the problem.

TABLE 1 SHOWING CERTAIN VARIATIONS BETWEEN INDIVIDUAL PLANTS OF WHEAT

Club-California No. 459					White Australian-California No. 452					Propo				
Cent No.	Number kernels in plant	Weight of kernels per plant (Grams)	Per cent nitrogen in kernels	Per cent total protein	Cent No.	Number kernels in plant	Weight of kernels per plant (Grams)	Per cent nitrogen in kernels	Per cent total protein	Cent No.	Number kernels in plant	Weight of kernels per plant (Grams)	Per cent nitrogen in kernels	Per cent total protein
4718	219	10.241	1.714	9.74	4813	103	3.7808	2.182	12.39	7317	144	7.0617	1.793	10.18
4704	244	10.538	2.614	14.85	4818	119	5.0306	1.835	10.42	7315	150	7.2607	1.864	10.09
4711	244	10.451	2.646	15.03	4821	121	5.0302	2.345	13.32	7316	150	5.9803	2.078	11.80
4722	246	11.143	1.979	7.26	4810	123	4.9826	1.740	9.88	7302	168	8.8266	1.944	11.04
4723	254	11.254	2.031	11.54	4809	128	5.1400	1.954	11.10	7310	173	8.1147	1.786	10.14
4701	263	12.480	2.232	12.68	4816	128	5.8740	1.732	9.84	7318	173	9.2274	1.852	10.52
4721	264	9.397	2.758	15.67	4814	135	5.5290	1.752	9.95	7320	199	9.5840	1.776	10.09
4707	266	11.797	2.359	13.40	4819	139	5.9616	1.609	9.14	7311	200	7.7578	1.855	10.54
4724	270	12.176	2.473	14.05	4820	144	6.1408	3.459	13.97	7303	210	10.7348	1.419	8.06
4702	272	12.480	2.232	12.74	4817	150	5.2002	2.002	11.47	7308	218	10.5668	1.547	8.79
4720	278	11.401	1.866	10.60	4823	151	6.6950	1.321	7.50	7314	219	11.4692	1.725	9.80
4716	281	13.061	1.889	10.73	4808	152	6.4767	1.794	10.19	7309	229	12.5288	1.779	10.10
4709	290	12.004	1.824	10.36	4806	155	6.9606	1.595	9.06	7312	245	10.7326	1.954	11.10
4713	294	12.054	1.907	10.59	4822	157	6.6148	2.696	1.31	7321	261	12.9620	2.415	13.72
4719	295	11.897	1.476	8.38	4811	159	6.3916	2.417	13.73	7313	284	12.1798	2.499	14.19
4705	296	12.274	2.614	9.03	4824	166	5.9324	2.109	11.95	7307	306	13.173	2.362	13.45
4712	299	13.104	2.485	14.12	4812	176	8.2980	1.722	9.78	7319	325	13.9718	2.312	13.13
4717	303	12.536	1.597	9.07	4807	182	8.6880	2.069	11.75	7306	338	13.0815	1.629	9.20
4715	304	13.003	1.995	11.33	4805	187	8.5149	1.830	10.39	7304	300	8.9246	1.946	11.05
4710	325	13.825	1.931	10.97	4815	196	8.1176	1.928	10.95	7305	310	7.7959	1.821	10.34
4708	347	13.850	2.141	12.16	4825	203	7.1600	1.979	11.24	.....	.....	.....	.....	.....
4714	376	17.074	2.010	11.42	4803	286	6.7326	2.133	12.12	.....	.....	.....	.....	.....
4706	412	17.119	2.856	16.22	4804	206	9.5090	1.770	10.05	.....	.....	.....	.....	.....
4703	539	24.388	1.254	7.12	4802	232	7.5580	1.713	9.73	.....	.....	.....	.....	.....
4725	555	24.672	1.432	8.13	4801	257	1.9316	1.704	9.69	.....	.....	.....	.....	.....
Av.	309	13.433	2.064	11.49	.....	163	6.7224	1.936	11.00	.....	244	10.0967	1.9178	10.89

## THE CONSTANCY OF CHANGE TOWARD STARCHINESS IN WHEAT KERNELS

The phase presented in this portion of the study has to do with the tendency of varieties toward a constancy of change from the type seeded. The central idea was that if there really existed a strong tendency toward the development of a distinctly starchy nature, under the conditions which here obtain, not only would the progeny of perfectly typical glutenous kernels show a considerable number of more or less starchy kernels and a lower protein (gluten) content, but spotted kernels would show a similar change in their progeny; and the entirely changed kernels, carrying 100 per cent of starchy kernels in the original, would show a still lower percent of typical kernels and a very low protein content in the progeny, remaining, in fact, essentially starchy.

The plan of the experiment which extended over the seasons of 1906-08, inclusive, was to grow a number of varieties of wheat at several points in the state, the physical and chemical condition of the originals used being first determined, and to follow the changes in the progeny in each instance during the season and the total period of the trials.

The experiments were conducted in the fields at Modesto, Ceres, and Tulare as typical of San Joaquin Valley conditions, and at Yuba City and Davis as representative of Sacramento Valley conditions. In order to follow better such changes as occurred, by the eye as well as by analysis, such varieties of wheat were selected as had typical kernels of distinctive appearance.

## THE NITROGEN OF TYPICAL vs. SPOTTED WHEAT KERNELS OF THE SAME VARIETY

At the outset a number of varieties of wheat were hand-separated into three primary lots or groups, based upon the physical appearance of the kernels, as follows:

Group I, consisting entirely of perfectly typical (glutenous) kernels;

Group II, consisting of kernels of the same varieties of wheat

which were of an entirely starchy character, showing no typical kernels whatsoever;

Group III, consisting of kernels of the same varieties of an intermediate appearance upon which the percentage of typical kernels was determined in each case.

Each of these lots was subjected to a determination of the nitrogen content in order to ascertain whether this distinctive appearance of the kernels was a fair indication of their nitrogen content.

The results are subtended:

#### NITROGEN CONTENT (DRY BASIS)

	Typical kernels	More or less starchy kernels
Sample no. 1	1.75	1.45
Sample no. 2	1.75	1.54
Sample no. 3	1.67	1.54
Sample no. 4	1.85	1.57
Sample no. 5	1.98	1.74
Sample no. 6	2.13	1.78
Sample no. 7	2.09	1.73
Sample no. 8	2.03	1.74
Sample no. 9	1.98	1.52
Sample no. 10	2.02	1.90
Sample no. 11	2.12	1.90
Sample no. 12	2.27	2.08
Sample no. 13	1.82	1.68
Sample no. 14	2.03	1.68
Sample no. 15	2.33	1.83
Sample no. 16	1.83	1.62
Average	1.97	1.69

These results seem to show that the physical appearance of the grain may, in general at least, be taken as a fair index of its relative gluten or protein content.

Each of the above described groups was drill-seeded in a number of rows in a uniform manner and under uniform soil conditions and given the same cultural treatment.

The results of the experiments in 1906 are tabulated below:

TABLE 2.—SHOWING PHYSICAL AND CHEMICAL DEVIATION OF WHEAT KERNELS FROM TYPE SEEDED IN 1906

MODESTO													
Lab. No.	Name	Condition of seed	Date of harvest	Per cent of typical kernels	Number in 10 kernels	As analyzed				Calculated to dry basis			
						Moisture	Total nitrogen	Protein	Ash	Total nitrogen	Protein	Ash	
644	Kubanka	Clear	Original	100.0	203	9.99	2.10	11.93	1.83	2.33	13.25	2.03	
709	Kubanka	.....	June 27	98.9	252	10.98	2.33	13.24	1.99	2.62	14.91	2.24	
647	Kubanka	Spotted	Original	0.0	206	10.21	1.81	10.27	1.88	2.01	11.46	2.09	
711	Kubanka	.....	June 27	89.4	216	10.87	2.31	13.16	1.98	2.59	14.75	2.22	
650	Kubanka	White	Original	0.0	235	9.45	1.66	9.41	1.84	1.83	10.43	2.03	
713	Kubanka	.....	June 27	96.9	195	10.95	2.23	13.68	1.89	2.50	14.20	2.12	
653	Chul	Dark	Original	100.0	.....	10.27	2.05	11.68	1.84	2.39	14.62	2.05	
715	Chul	.....	June 12	94.1	219	11.96	1.81	10.36	2.00	2.05	11.69	2.25	
655	Chul	Spotted	Original	0.0	215	10.67	2.00	11.40	1.90	2.24	12.77	2.20	
717	Chul	.....	June 12	91.6	218	1.13	1.96	11.16	2.00	2.21	12.56	2.25	
338A	Turkey Red	Clear	Original	100.0	320	12.74	1.59	9.05	1.74	1.82	10.37	1.99	
338A3	Turkey Red	.....	June 27	98.0	314	11.28	2.08	11.80	2.08	2.34	14.34	2.34	
338B	Turkey Red	Spotted	Original	0.0	310	12.34	1.42	8.09	1.72	1.62	9.23	1.96	
338B3	Turkey Red	.....	June 27	92.2	309	11.19	1.98	11.24	2.02	2.23	12.71	2.27	
200	Kubanka	Spotted	Original	38.3	250	13.56	2.13	12.14	1.88	2.47	14.04	2.18	
725	Kubanka	.....	June 21	97.9	246	11.01	1.96	11.16	1.88	2.20	12.51	2.11	
505	Marouani	Spotted	Original	43.2	216	11.36	1.70	9.68	1.74	1.94	11.06	1.96	
725	Marouani	.....	July 14	95.3	171	11.50	2.15	12.20	1.98	2.43	13.71	2.24	
573	Turkey Red	Clear	Original	100.0	315	14.65	2.04	11.64	1.94	2.39	13.39	2.27	
733	Turkey Red	.....	June 27	99.3	306	10.33	2.16	12.28	2.02	2.40	13.63	2.25	

TABLE 2.—SHOWING PHYSICAL AND CHEMICAL DEVIATION OF WHEAT KERNELS FROM TYPE SEEDED IN 1906—(Concluded)

Lab. No.	Name	Condition of seed	Date of harvest	Per cent of typical kernels	Number of kernels in 10 grains	As analyzed				Calculated to dry basis		
						Moisture	Total nitrogen	Protein	Ash	Total nitrogen	Protein	Ash
644	Kubanka	Clear	Original	100.0	203	9.99	2.10	11.93	1.83	2.33	13.25	2.03
645	Kubanka	.....	July 2	55.6	217	10.11	1.92	10.91	2.04	2.14	12.12	2.26
647	Kubanka	Spotted	Original	0.0	206	10.21	1.81	10.27	1.88	2.03	11.56	2.09
648	Kubanka	.....	July 2	20.8	197	10.38	1.82	10.34	1.98	2.03	11.56	2.21
650	Kubanka	White	Original	0.0	235	9.45	1.66	9.41	1.84	1.83	10.42	2.03
651	Kubanka	.....	July 2	63.5	205	12.79	1.70	9.72	2.16	1.95	11.15	2.48
653	Chul	Dark	Original	100.0	210	10.27	2.05	11.68	1.84	2.39	14.62	2.05
645	Chul	.....	June 30	59.4	203	10.50	1.81	10.28	1.99	2.03	11.56	2.22
656	Chul	Spotted	Original	0.0	215	10.67	1.73	9.80	1.90	1.93	11.00	2.11
657	Chul	.....	June 30	76.0	221	10.31	1.94	11.02	2.00	2.17	12.36	2.12
659	Chul	Light	Original	0.0	216	10.42	1.44	8.17	1.77	1.61	9.13	2.99
660	Chul	.....	June 30	52.0	220	12.49	1.87	10.64	2.00	2.12	12.04	2.28
338A	Turkey Red	.....	Original	100.0	330	12.74	1.59	9.05	1.74	1.71	9.71	1.99
338A1	Turkey Red	Clear	Original	65.3	298	10.41	1.77	10.05	1.87	1.98	11.25	2.09
338B	Turkey Red	.....	July 7	0.0	310	12.34	1.42	8.09	1.72	1.62	9.20	1.96
338B1	Turkey Red	Spotted	Original	67.3	302	10.15	1.77	10.03	1.97	1.97	11.20	2.19
677	Kubanka	.....	Original	23.7	232	10.78	1.50	8.59	1.84	1.68	9.56	2.06
678	Kubanka	Spotted	July 2	63.4	230	10.31	1.91	10.84	2.05	2.13	12.04	2.29
671	Kahla	.....	Original	75.2	208	10.34	2.15	12.20	2.17	2.40	13.55	2.42
672	Kahla	Spotted	July 5	53.0	207	10.04	2.06	11.76	2.19	2.30	13.11	2.43
666	Kharkov	.....	Original	93.1	301	10.47	2.13	12.10	2.15	2.38	13.68	2.40
861	Kharkov	Spotted	July 5	83.9	310	11.64	1.89	11.75	2.07	2.18	12.44	2.40
862	Beloglina	.....	Original	59.9	310	10.49	1.81	10.28	2.00	2.02	11.48	3.56
863	Beloglina	Spotted	June 23	77.7	314	12.72	1.83	10.40	2.22	2.09	11.91	2.54

YUBA CITY

## DISCUSSION OF 1906 RESULTS

Examination of the 1905-06 results shows that in physical appearance none of the seven originals carrying 100 per cent of typical kernels (Group I) maintained their perfection in their progeny—all showed some starchy kernels. The fact that those grown at Modesto showed 97.5 per cent of typical kernels in their progeny while those grown at Yuba City showed but 60.1 per cent very strongly suggests there must have been some local condition of either soil or climate which influenced this change.

The average protein content of this group at Modesto and Yuba City decreased slightly, as did the percentage of typical kernels, showing 13.15 per cent total protein in the originals and 12.91 per cent in the progeny; while at Yuba City the results were 12.52 per cent for the originals and 11.64 for the progeny. It will be noted also that the Modesto samples possessed somewhat higher total protein as well as a larger percentage of typical kernels than did those grown at Yuba City.

Under Group II at Modesto the progeny showed an average of 92.5 per cent of typical kernels from originals carrying absolutely none, and a comparative protein content of 10.97 per cent for the originals against 13.55 per cent for the progeny, or an increase of 2.58 per cent.

Group III at Modesto carried an average of 40.7 per cent typical kernels in the originals and 96.6 per cent in the progeny, and a protein content of 12.55 and 13.11 respectively, a distinct increase in both respects. At Yuba City this group showed 62.9 and 69.5 per cent typical kernels in the originals and progeny respectively, and a protein content of 12.07 per cent against 11.92 per cent.

Considered as a whole the results are expressed below:



	Group I			Group II			Group III		
	Number of samples in group	Per cent typical kernels	Per cent total protein	Number of samples in group	Per cent typical kernels	Per cent total protein	Number of samples in group	Per cent typical kernels	Per cent total protein
1906									
Original .....	7	100.0	12.74	9	0.0	10.58	6	55.6	12.23
Progeny .....	7	81.5	12.78	9	75.6	12.50	6	78.5	12.32
					General average per cent typical kernels	General average per cent total protein			
1906				Total number of trials					
Original .....				22	47.39	11.71			
Progeny ... ..				22	78.2	12.54			

It is here shown that the average per cent of typical kernels in all the originals was 47.4 and in the progeny 78.2, and the protein content 11.71 per cent and 12.54 per cent respectively. *It is perfectly evident from these figures that a wheat which consists of 100 per cent starchy kernels may, in a single season, revert to practically a perfect wheat, or to essentially the same average protein content for the season and region as a wheat grown from perfectly glutenous kernels.*

Instances of this are particularly shown in samples no. 713 and no. 725. In the first the original consisted of 100 per cent of entirely starchy kernels and still produced 96.9 per cent of typical kernels in the progeny. In the second the same is shown to a somewhat smaller extent, the original carrying but 38.3 per cent typical kernels and still producing 97.9 per cent in the progeny.

*Other trials.*—Trials were also made to determine the percentage of typical seed in six varieties grown at Modesto from seed in which the percentage of typical kernels had been previously determined, using 1000 kernels as a basis. These were also seeded in rows and were harvested in the hard dough stage. Counts were then made to determine the percentage of typical kernels in the progeny. The results appear below.

Name	Per cent of typical kernels in original 1905	Per cent of typical kernels 1906
Frete's	94.0	95.2
Redwinter	51.0	91.5
Hungarian	78.7	95.3
Kubanka	38.3	72.6
Koola	16.8	93.9
Marouani	43.2	93.1
Average	53.7	90.2

In each of the above cases the percentage of typical kernels increased in the 1906 crop over those of the 1905 crop and the average increase was from 53.7 to 90.2; it seems probable that some climatic or soil factor is more than likely to have been the cause of this change. This is particularly noticeable upon comparing the results between the two stations.

The above figures not only do not indicate that there was any strongly marked tendency toward lowering in quality, but, on the other hand, the general tendency seems to be upward for this season, particularly at Modesto.

*The Relation of Moisture.*—It has been shown repeatedly by many investigators that the composition of plants varies considerably in different localities and in different seasons, and that the principal factor seems to be the climate instead of the soil, variations due to the latter being very slight if any. This feature will be presented more in detail later in this paper. While the relation of the water content of the soil at various stages of the plant growth to the protein content of wheats also forms a portion of these investigations for later presentation, it may be said that the Utah Station has conducted a number of experiments upon the effect of water upon the composition of plants, the method of which in each case has been to apply different amounts of water throughout the season on contiguous plats of uniform land. The following selected results from the Utah experiments as affecting the protein content of the grain are of interest in this connection.

Inches of water applied	Protein content of		
	Corn kernels	Oat kernels	Wheat kernels
7.5	15.08	20.79	26.72
15.0	13.48	17.29	19.99
37.3	12.52	15.49	16.99

It is shown here that as the amount of water is increased the protein content decreases.

Others have also observed the effect of water upon the composition of crops. Mayer, in Holland, showed that on a soil having 10 per cent of water the crop contained 10.6 per cent of protein, while on a soil with 30 per cent of water the protein percentage was only 6.6. Carleton calls attention to the fact that in the same varieties of wheat grown in the humid and arid regions of the United States the protein content was 11.94 per cent for the former and 14.4 per cent for the latter. Experiments conducted at Rothamstead, England, show that barley in a wet year contained 9.81 per cent of protein and in a dry year 12.99 per cent.

The particular connection of this review of the observation of other workers to the results here presented lies in the differences in the results secured in 1906 between the Modesto and the Yuba City stations, at the former of which both the percentage of typical kernels and the protein content in the progeny was distinctly higher than at the latter. These three factors are contrasted below.

	Inches rainfall Dec. 1-June 30	Per cent typical kernels	Per cent total protein
Modesto	11.94	95.35	13.50
Yuba City	26.68	63.99	11.74

A very casual observation of these results shows that both the distinct difference in percentage of typical kernels and that between the protein content of the grain of the two stations was without doubt very strongly, if not entirely, determined by the difference in moisture which the grain received after planting.

#### EXPERIMENTS OF 1907

In 1907 the experiment described above was continued at Modesto and Yuba City, and also extended to Tulare. The results are stated below.

TABLE 3.—SHOWING PHYSICAL AND CHEMICAL DEVIATION OF WHEAT KERNELS FROM TYPE SEEDED IN 1907

Lab. No.	Name	Condition of seed	Date of harvest	MODESTO		As analyzed				Calculated to dry basis			
				Per cent of typical kernels	Number of kernels in 10 grams	Moisture	Total nitrogen	Protein	Ash	Total nitrogen	Protein	Ash	
645C	Kubanka	Clear	Original	100.0	217	11.10	1.74	9.96	2.04	1.96	11.20	2.21	
927A	Kubanka	.....	June 21	48.0	252	11.95	1.71	9.74	1.76	1.94	11.06	1.98	
928C	Kubanka	Spotted	Original	0.0	221	11.18	1.48	8.37	1.69	1.65	9.42	1.90	
928A	Kubanka	.....	June 21	49.0	249	11.50	1.70	9.66	1.74	1.91	10.92	.....	
929C	Kubanka	White	Original	0.0	267	11.92	1.24	7.09	1.58	1.41	8.05	1.96	
929A	Kubanka	.....	June 21	56.0	242	11.52	1.69	9.61	1.83	1.89	10.87	2.07	
926C	Chul	Dark	Original	100.0	232	11.46	1.94	11.06	1.75	2.19	12.48	1.97	
926A	Chul	.....	June 12	60.0	227	11.61	1.71	9.74	2.04	1.93	11.02	2.31	
925C	Chul	Spotted	Original	0.0	223	11.50	1.65	9.45	2.00	1.87	10.67	2.26	
925A	Chul	.....	June 12	42.0	220	11.93	1.41	8.05	1.83	1.60	9.14	2.07	
924C	Chul	Dark	Original	0.0	232	11.57	1.71	10.75	1.81	2.12	12.14	2.04	
924A	Chul	.....	June 12	49.0	242	11.89	1.52	8.71	1.88	1.73	9.88	2.11	
923C	Kharkov	Clear	Original	100.0	.....	11.76	2.03	11.56	2.14	2.30	13.10	2.42	
923A	Kharkov	.....	.....	93.0	295	11.80	2.12	12.08	1.96	2.40	13.69	2.22	
930C	Fretes	Spotted	Original	83.3	145	11.73	1.69	9.68	2.03	1.92	10.96	2.29	
930A	Fretes	.....	June 21	85.1	235	11.99	.....	.....	.....	.....	.....	.....	
931C	Erivan	Spotted	Original	80.9	297	11.26	2.06	11.76	1.89	2.32	13.26	2.13	
931A	Erivan	.....	June 21	76.7	365	11.56	1.61	9.21	1.95	1.83	10.42	.....	
932C	Gharnovka	Spotted	Original	21.4	263	11.52	1.55	8.82	1.89	1.63	9.28	2.13	
932B	Gharnovka	.....	June 21	29.5	267	12.07	1.59	9.07	1.68	1.81	10.32	1.91	
933C	Koola	Spotted	Original	87.0	188	13.59	1.59	11.54	1.93	2.34	13.36	2.23	
933A	Koola	.....	June 21	57.1	255	11.88	1.80	10.28	1.91	2.04	11.66	2.16	
934C	Kubanka	Spotted	Original	78.0	200	11.47	1.64	9.39	2.00	1.86	10.61	2.26	
934A	Kubanka	.....	June 21	40.9	225	11.75	1.64	9.34	1.74	1.85	10.59	1.97	
935C	Velvet Don	Spotted	Original	44.1	275	11.46	1.58	9.00	1.69	1.71	10.17	1.91	
935A	Velvet Don	.....	June 21	37.3	280	11.78	1.80	10.28	1.77	2.04	11.65	2.00	

TABLE 3.—SHOWING PHYSICAL AND CHEMICAL DEVIATION OF WHEAT KERNELS FROM TYPE SEEDED IN 1907—(Continued)

Lab. No.	Name	Condition of seed	Date of harvest	YUBA CITY		As analyzed				Calculated to dry basis		
				Per cent of typical kernels	Number of kernels in 10 grams	Moisture	Total nitrogen	Protein	Ash	Total nitrogen	Protein	Ash
645A	Kubanka	Clear	Original	100.0	204	12.53	2.36	13.46	1.80	2.70	15.39	2.06
647	Kubanka	.....	July 2	48.6	201	13.24	1.68	9.58	1.71	1.93	11.03	1.97
648S	Kubanka	Spotted	Original	0.0	200	12.71	1.69	9.64	2.04	1.94	11.05	2.33
850	Kubanka	.....	July 2	48.3	203	13.08	1.61	9.20	1.67	1.85	10.58	1.92
651W	Kubanka	White	Original	0.0	205	12.79	1.70	9.72	2.16	1.94	11.15	2.48
853	Kubanka	.....	July 2	38.9	224	13.50	1.59	9.07	1.98	1.84	10.48	2.29
654	Chul	Dark	Original	100.0	203	10.50	1.81	10.28	1.99	2.02	11.49	2.22
855	Chul	.....	June 29	58.1	202	12.98	1.48	8.47	2.01	1.71	9.74	2.31
661	Chul	White	Original	0.0	201	12.96	1.93	10.03	1.89	1.89	11.54	2.17
857	Chul	.....	June 29	82.8	200	13.88	1.18	6.73	1.85	1.37	7.81	2.15
338A1	Turkey Red	Clear	Original	0.0	298	10.41	1.77	10.05	1.87	1.97	11.22	2.13
338A5	Turkey Red	.....	July 13	75.7	310	11.86	2.86	16.32	1.77	2.81	18.52	2.01
666	Kharkov	Spotted	Original	93.1	301	10.47	2.13	12.10	2.15	2.38	13.52	2.40
859	Kharkov	.....	Aug. 17	83.9	310	11.64	1.71	10.75	2.07	2.18	12.44	2.40
669	Weissenberg	Spotted	Original	87.7	304	10.04	2.09	11.88	2.18	2.32	13.32	2.42
862	Weissenberg	.....	Aug. 16	89.1	270	13.67	2.43	13.87	3.39	3.39	16.06	2.38
663	Fretes	Spotted	Original	100.0	217	10.10	2.02	11.48	2.10	2.25	12.80	2.34
866	Fretes	.....	July 2	83.5	225	12.85	1.72	9.82	2.12	1.94	11.27	2.43
678	Kubanka	Spotted	Original	63.4	215	10.31	1.91	10.84	2.05	2.13	12.12	2.29
869	Kubanka	.....	July 8	89.2	184	13.36	3.59	20.48	1.93	4.16	23.64	2.22
684	Velvet Don	Spotted	Original	79.5	217	10.05	2.02	11.98	1.95	2.25	13.74	2.17
870	Velvet Don	.....	July 2	90.1	239	12.84	2.13	12.17	2.27	2.45	13.96	2.61
696	Gharnovka	Spotted	Original	6.5	208	11.28	1.55	8.80	2.02	1.74	9.92	2.27
872	Gharnovka	.....	July 2	66.6	210	12.06	1.90	10.84	1.74	2.11	12.05	.....
874	Belogina	Spotted	Original	59.9	305	10.50	1.66	9.41	1.95	1.85	9.43	2.21
682	Belogina	.....	July 23	77.4	314	12.72	1.82	10.40	2.22	2.09	11.91	2.54

TABLE 3.—SHOWING PHYSICAL AND CHEMICAL DEVIATION OF WHEAT KERNELS FROM TYPE SEEDED IN 1907—(Concluded)

Lab. No.	Name	Condition of seed	Date of harvest	Per cent of typical kernels	Number of kernels in 10 grams	As analyzed				Calculated to dry basis		
						Moisture	Total nitrogen	Protein	Ash	Total nitrogen	Protein	Ash
681A	Beloglina	Spotted	Original	66.1	268	11.03	1.78	10.11	2.03	1.99	11.36	2.28
892	Beloglina	.....	June 21	59.4	271	13.21	1.94	9.51	2.04	1.92	10.96	2.22
684	Velvet Don	Spotted	Original	79.4	217	10.05	2.02	11.98	1.95	2.41	13.74	2.17
894	Velvet Don	.....	June 6	93.5	249	13.31	1.42	8.10	1.97	1.64	9.34	2.04
896	Kharnovka	Spotted	Original	6.5	298	11.28	1.55	8.80	2.02	1.74	9.92	2.27
899	Kharnovka	.....	June 11	44.1	303	12.07	1.53	8.75	1.97	1.97	10.71	2.24
648	Kubanka	Spotted	Original	50.8	200	12.71	1.69	9.64	2.04	1.94	11.05	2.33
903	Kubanka	.....	July 29	20.0	225	13.34	1.61	9.18	1.68	1.87	10.60	1.93
338B2	Turkey Red	Spotted	Original	50.1	289	10.41	1.78	10.11	1.94	1.96	11.28	2.18
902	Turkey Red	.....	July 29	66.8	283	14.44	1.75	10.01	1.82	2.05	11.71	2.12
664	Fretes	Typical	Original	100.0	213	10.37	2.06	11.70	2.05	2.29	13.05	2.28
904	Fretes	.....	June 3	0.0	220	13.87	1.61	9.17	1.86	1.86	10.60	2.16
676	Kharnovka	Spotted	Original	9.0	228	11.09	1.56	8.86	2.01	1.75	9.95	2.26
900	Kharnovka	.....	July 29	46.8	232	12.97	1.76	10.00	2.07	2.11	12.06	2.37

*Discussion of 1907 Results.*—Essentially the same results are shown here as in the preceding season. Of the seven (7) originals in Group I, all of the progeny dropped distinctly below the original standard, showing 55.99 per cent of typical kernels, while the eight samples in Group II, carrying distinctly starch kernels in the originals, all increased by about the same amount as the others dropped, the average per cent of typical kernels in the progeny being 55.2 per cent, or less than 0.7 per cent below those of Group I.

In the group of typical kernel originals, only one out of seven increased in protein content, the average per cent in the originals being 12.79 against 11.20 per cent in the progeny, or a drop of 1.59 per cent. In Group II, consisting entirely of the distinctly starchy kernels, there was a universal increase in the typical kernels in the progeny, and three out of eight increased in protein content, the averages being 10.65 per cent for the originals and 11.02 per cent for the progeny. Group III showed 58.1 per cent typical kernels in the originals and 64.1 per cent in the progeny, an increase of 6 per cent; and 11.50 and 12.25 per cent total protein, respectively.

Summarizing by groups as in the results of the preceding year, the figures appear as below:

	Group I			Group II			Group III		
	Number in group	Per cent typical kernels	Per cent total protein	Number in group*	Per cent typical kernels	Per cent total protein	Number in group	Per cent typical kernels	Per cent total protein
1907									
Original .....	7	100.0	12.79	8	0.0	10.65	18	58.10	11.50
Progeny .....	7	55.9	11.24	8	55.2	11.02	18	64.10	12.25
					General average per cent typical kernels	General average per cent protein			
1907				Total number trials					
Original .....				33	58.1	11.56			
Progeny .....				33	60.2	11.72			

Eighteen of these thirty-three samples showed an increase of typical kernels in the progeny over the originals, the respective average being 60.2 and 54.6. The protein content increased in sixteen (16) out of thirty-three cases, with averages for originals and progeny of 11.56 per cent against 11.72 per cent.

#### EXPERIMENTS OF 1908

The same experiments were continued in 1908 at Davis instead of Yuba City, and at Ceres instead of Modesto. The tabulated results of this season are presented below:





TABLE 4.—SHOWING PHYSICAL AND CHEMICAL DEVIATION OF WHEAT KERNELS FROM TYPE SEEDED IN 1908—(Continued)

Lab. No.	Name	Condition of seed	Per cent of typical seed	Date of harvest	Per cent typical seed	Number kernels in 10 grams	As analyzed				Calculated to dry basis					
							Moisture	Total nitrogen	Gliadin	Ash	Total nitrogen	Total protein	Gliadin	Ash		
919/08	Hybrid	.....	100.0	July 16	100.0	377	12.74	2.27	12.98	5.13	1.76	.....	2.61	14.87	.....	.....
851	Kubanka	Spotted	42.8	Original	42.8	210	13.09	1.75	10.00	3.09	1.62	.....	2.02	11.51	3.55	1.87
851/08	Kubanka	.....	88.4	June 27	88.4	.....	11.35	1.99	11.33	3.41	1.54	.....	2.24	12.78	.....	.....
338B5	Turkey	Spotted	74.0	Original	74.0	313	11.16	1.94	11.11	3.77	1.77	.....	2.19	12.50	4.24	1.99
338B5/08	Turkey	.....	100.0	July 16	100.0	.....	12.14	2.38	13.52	4.78	1.83	.....	2.69	15.36	.....	.....
745B	Kubanka	Spotted	66.2	Original	66.2	.....	13.57	1.58	9.01	3.43	1.80	.....	2.69	10.42	.....	.....
745B/08	Kubanka	.....	100.0	July 5	100.0	215	12.13	2.39	13.82	4.09	1.77	.....	2.76	15.73	.....	.....
872	Gharovka	Spotted	66.6	Original	66.6	210	12.06	1.90	10.84	3.08	1.74	.....	2.22	12.65	3.50	.....
872/08	Gharovka	.....	98.4	July 1	98.4	215	12.93	2.42	13.81	4.16	1.75	.....	2.76	15.72	.....	.....
962	Fife	Clear	100.0	Original	100.0	315	8.64	1.96	11.18	.....	.....	.....	2.15	12.23	.....	.....
962/08	Fife	.....	100.0	July 7	100.0	319	12.15	2.61	14.88	5.13	1.72	.....	2.97	16.94	.....	.....
868	Kubanka	Spotted	89.18	Original	89.18	184	13.36	3.59	20.48	4.05	1.93	.....	4.15	23.64	4.67	2.22
868/08	Kubanka	.....	99.70	July 2	99.70	192	13.99	2.42	13.79	4.28	1.58	.....	2.79	15.91	.....	.....
847	Kubanka	Spotted	48.6	Original	48.6	.....	13.24	1.68	9.58	3.37	1.71	.....	1.93	11.04	.....	.....
847/08	Kubanka	.....	98.5	July 1	98.5	201	13.81	2.19	12.47	4.00	1.56	.....	2.53	14.46	.....	.....
870	Velvet Don	Spotted	90.1	Original	90.1	239	12.84	2.13	12.17	4.12	2.27	.....	2.44	13.96	.....	.....
870/08	Velvet Don	.....	99.1	July 2	99.1	214	13.36	2.31	13.17	4.52	1.54	.....	2.66	15.20	.....	.....



## GENERAL DISCUSSION AND CONCLUSION

Reviewing the results of the three years, the general average of typical kernels in the original was 63.63 per cent and of the seed produced therefrom 77.99 per cent, while the protein content was 12.34 per cent and 12.95 per cent respectively.

The results as a whole show:

First—That in general the physical appearance of durum and red wheats is a fair indication of their relative protein content; kernels having a distinctly horny or glutenous appearance being higher in protein than those of a more or less dull or starchy appearance.

Second—That there is a wide seasonal fluctuation in protein content of wheat which may become so great as to overbalance almost entirely any hereditary tendency of starchy originals to produce the same characteristics in their progeny.

Third—That the protein content of wheat in a locality is undoubtedly largely dependent upon the seasonal precipitation in such locality.

Fourth—That the use of perfectly typical glutenous seed is invariably followed under California conditions by a lowering of the gluten content, as indicated both by the physical appearance of the grain and by its protein content.

Fifth—That if the original carries a considerable percentage of starchy kernels the progeny usually shows an increase toward the typical character to a degree determined by the character of the season in the locality. This is especially so with reference to the precipitation, which in some instances may have such a strong influence as to cause a practically perfect grain to result from an original seed carrying 100 per cent of starchy kernels.

Sixth—The last tabulation by groups further indicates quite strongly, however, that as a matter of fact the character of the seed used has quite a marked influence upon the progeny, and that the quality of the seed used, to some degree at least, determines the character of the resultant crop, for it will be noted that as the originals decrease in both percentage of typical kernels and protein the progeny in each case decrease in the same order, although the effect of this is materially lessened and sometimes

almost entirely overcome by the character of the season, as shown by the other results.

### THE RELATION OF THE TIME OF PLANTING TO THE PROTEIN CONTENT OF WHEAT KERNELS

On account of certain differences in the physical appearance of kernels from early and late planted wheats in preceding seasons, experiments were conducted in 1908 to ascertain more definitely the relation of early and late planting to the protein content of wheat kernels. In these experiments it was intended to conduct parallel series of plantings at the University Farm, Davis, and at the substation at Tulare. At the former place, however, conditions were not favorable for early seeding, and all had to be planted too late to be at all satisfactory in presenting any decided contrast in planting time, particularly for the winter wheats, there being but fifteen days between the early and late seeding. The first plantings of the season were not made until February 27, and the second plantings on March 13. At Tulare there was a wider difference, the plantings having been made on December 12, and January 17.

As a matter of record, however, the analyses of these grains are presented below:

TABLE 5.—SHOWING THE RELATION OF TIME OF PLANTING TO THE PHYSICAL AND CHEMICAL CHARACTERISTICS OF WHEAT KERNELS

Lab. No.	First planting				Second planting			
	Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent	Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent
2/05	100.0	11.81	1.41	1.76	67.1	10.67	2.40	2.04
847A	68.3	11.19	1.61	1.76	40.6	10.62	2.19	2.03
870A	100.0	13.78	1.80	1.73	100.0	13.74	3.12	1.72
873A	100.0	13.38	2.32	1.73	84.0	13.08	3.17	1.72
909A	96.9	14.41	3.60	2.07	97.7	14.89	4.99	1.49
910A	96.2	12.99	6.88	1.93	60.2	9.77	1.80	2.03
911D	95.6	13.71	4.16	2.06	99.4	18.26	8.05	2.37
917D	100.0	17.25	6.95	2.16	100.0	19.68	5.41	2.08
919C	85.7	14.87	5.88	2.01	99.4	17.27	5.82	2.22
922A	99.0	14.58	4.15	2.00	99.1	12.07	3.09	2.10
595	95.3	14.91	5.90	1.89	93.3	13.24	5.53	2.13
Av.	94.2	13.88	4.06	1.92	85.5	13.94	4.14	1.99

## TULARE, 1908

Lab. No.	First planting				Second planting			
	Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent	Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent
901	.....	10.34	3.58	.....	.....	11.30	4.75	.....
896	14.3	9.82	3.09	1.92	85.1	12.95	4.32	1.68
870	20.7	9.94	3.43	1.88	68.1	12.15	4.97	1.87
848	30.8	10.96	3.67	2.03	60.1	11.53	4.04	1.84
892	74.5	11.41	3.51	2.04	98.1	13.63	4.48	2.04
338A6	78.3	11.47	3.60	1.92	97.4	13.52	4.44	2.08
Av.	43.7	10.65	3.48	1.96	81.7	12.51	4.50	1.90
Grand av.	68.9	12.26	3.77	1.94	83.6	13.22	4.32	1.95

For the reason indicated above the plantings made at Davis should not be regarded as having much bearing upon the question. Considering the plantings made at Tulare, however, it will be noted that in every case the late plantings showed both a higher percentage of typical kernels and a higher protein and gliadin content than the early plantings.

The average in the case of the Tulare samples was:

	Number of trials	Per cent typical kernels	Per cent total protein	Per cent gliadin
Early plantings .....	5	43.7	10.65	2.48
Late plantings .....	5	81.7	12.51	4.50

Further trials were made during the season of 1909-10 at Davis and Tulare. The planting dates at these stations were as follows:

	Early	Late
Davis	February 27th	March 13th
Tulare	December 26th	January 25th

TABLE 6 SHOWING THE RELATION OF TIME OF PLANTING TO THE PHYSICAL AND CHEMICAL CHARACTERISTICS OF WHEAT KERNELS

Lab. No.	First planting				Second planting			
	Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent	Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent
870B	98.5	15.77	5.77	1.75	99.1	15.46	6.16	1.90
873B	94.8	16.14	6.00	1.82	97.8	15.86	6.49	1.88
847C	99.0	15.46	6.05	1.73	98.0	16.01	6.52	2.13
909B	55.0	17.62	8.16	1.99	58.2	18.02	7.87	2.75
910B	97.9	16.66	6.20	1.79	100.0	16.82	5.94	1.98
922B	99.0	18.34	8.01	1.80	99.1	18.42	5.68	1.89
917E	98.0	18.42	8.35	2.21	100.0	20.02	7.44	2.45
388a5	100.0	18.18	7.71	2.17	100.0	18.58	6.92	2.28
875B	100.0	18.42	8.18	2.02	100.0	19.20	8.20	2.39
911E	.....	16.82	8.11	1.79	.....	19.44	9.14	2.12
912D	100.0	20.00	8.88	2.28	100.0	18.66	6.39	2.48
914D	98.0	17.62	7.86	2.03	100.0	17.62	7.91	2.45
Av.	94.5	17.45	7.44	1.94	95.6	17.84	7.05	2.22

TABLE 6—(Continued)

Lab. No.	First planting				Second planting			
	Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent	Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent
1043aG	77.0	10.73	3.45	2.23	100.0	17.26	6.31	2.86
1040aG	48.0	11.13	3.75	2.24	100.0	17.21	5.50	2.20
1041aG	44.0	11.07	3.79	2.14	99.0	15.90	5.35	2.40
1042aG	89.1	13.12	4.99	.....	.....	18.91	6.65	2.59
1040aS	35.0	11.36	4.11	2.29	100.0	13.73	5.91	2.22
1041aS	66.0	10.50	3.68	2.10	100.0	10.33	.....	2.34
1042aS	100.0	12.89	5.06	2.06	.....	16.47	6.19	2.54
1043aS	53.6	10.62	3.96	2.37	100.0	16.47	.....	2.74
1151	99.6	15.38	7.32	.....	99.2	15.34	6.50	.....
1155	100.0	17.20	6.95	1.81	100.0	17.24	5.38	2.25
Av.	71.2	12.40	4.71	2.15	99.8	15.88	5.97	2.46
Grand av.	82.8	14.92	6.07	2.04	97.7	16.86	6.51	2.34

These results show that out of twenty-two (22) cases, seventeen (17) carried a higher protein content in the later plantings than in those planted early and that a like number out of the twenty-two carried either a larger percentage or an equal number of typical kernels in the late plantings.

The averages are shown below:

	Trials	Typical kernels	Total protein	Gliadin
Early plantings .....	22	82.8	14.92	6.07
Late plantings .....	22	97.7	16.86	6.51

To further secure data upon the effect of the time of planting upon the quality of the grain, twelve (12) varieties of wheat were seeded at Ceres on four different dates, in rows 16½ feet long and 12 inches apart, as follows:

First planting .....	November 28, 1908
Second planting .....	December 12, 1908
Third planting .....	December 31, 1908
Fourth planting .....	January 19, 1909

The grain made a good stand and grew well during the season and matured as shown in the following table:

Lab. No.	DATE OF RIPENING			
	Date of plantings			
	Nov. 28	Dec. 12	Dec. 31	Jan. 19
914	June 15	June 18	June 22	July 17
909	June 14	June 18	June 20	July 7
910	June 18	June 20	June 20	June 24
911	June 16	June 20	June 26	July 7
915	June 20	June 24	July 3	July 7
913	June 16	June 20	June 24	July 7
917	June 15	June 16	June 19	July 7
916	June 11	June 18	June 22	July 7
919	June 16	June 20	June 24	July 7
912	June 16	June 18	June 22	July 7
1895	June 18	June 20	June 22	June 24
2/05	June 15	June 15	June 15	June 15

This series of plantings is of special interest in that out of the twelve varieties planted all but one show the highest protein content in the latest planting, and the one exception shows the highest in the third planting. In this case the difference between the protein content of the third and fourth plantings is less than one-half of 1 per cent. It is further of interest to note that there was quite a regular increase in the protein content in the order of planting. While the season was evidently one conducive to the development of a relatively high quality of grain



generally, it is noticeable that the average percentage of typical kernels also follows the same order as the protein content, although the difference is slight. If the average be made after discarding the last sample, which constitutes the single exception above referred to, the order in the average of typical kernels will appear absolutely the same as that of the protein content, and very consistently bears out the results secured at Davis and Tulare, previously discussed. While it may reduce the yield, *relatively late seeding tends to produce a grain of better quality than does early planting.*

This same fact is further evidenced by a series of thirty-seven (37) types of New South Wales hybrid stocks seeded at Davis in the season of 1910-11; the dates of planting being December 12, 1910, and February 20, 1911. The analyses from this lot are tabulated below:

TABLE 7.—SHOWING THE RELATION OF THE TIME OF PLANTING TO THE PHYSICAL AND CHEMICAL CHARACTERISTICS OF WHEAT KERNELS  
CERES, 1909

Lab. No.	First planting					Second planting					Third planting					Fourth planting				
	Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent		Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent		Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent		Typical kernels Per cent	Total protein Per cent	Gliadin Per cent	Ash Per cent	
914	98.0	15.96	6.70	1.97		100.0	16.98	6.87	1.90		100.0	16.81	6.86	2.05		100.0	17.27	7.13	2.32	
909	100.0	15.73	7.21	1.80		100.0	16.70	7.78	2.15		100.0	17.15	7.55	2.17		100.0	17.89	7.38	2.12	
910	100.0	13.74	4.09	1.97		100.0	14.54	4.77	1.91		99.0	16.30	5.27	2.00		97.0	16.76	4.78	2.00	
911	97.0	14.54	5.27	1.81		98.0	16.30	6.45	1.87		100.0	16.87	6.46	2.11		100.0	17.49	5.88	1.85	
915	100.0	16.36	5.22	2.00		100.0	16.64	5.05	1.98		100.0	17.21	6.15	2.08		100.0	17.72	6.01	2.28	
913	100.0	17.38	6.86	1.79		100.0	17.89	6.83	1.89		100.0	18.29	6.56	1.99		100.0	19.06	6.57	2.15	
917	97.0	15.85	5.22	1.87		99.0	16.19	5.58	1.88		100.0	16.53	6.07	1.94		100.0	18.40	5.83	2.06	
916	100.0	17.04	6.07	1.85		100.0	17.61	6.37	1.89		100.0	17.44	6.13	2.08		100.0	19.03	6.65	2.28	
919	98.0	16.70	5.28	1.95		99.0	15.56	5.63	1.62		100.0	15.28	5.19	1.74		100.0	18.35	5.82	2.02	
912	85.0	17.72	6.37	1.90		100.0	18.40	6.82	2.01		100.0	18.35	6.95	2.11		100.0	18.63	6.42	2.27	
1595	100.0	16.13	5.21	2.14		100.0	16.13	5.29	2.23		100.0	16.59	5.39	2.16		100.0	17.10	5.32	2.20	
2/05	94.0	17.27	4.54	2.15		98.0	17.72	5.80	2.10		98.0	18.52	5.70	2.10		97.0	18.12	5.99	2.12	
Av.	97.3	16.20	5.67	1.93		99.5	16.72	6.10	1.95		99.8	17.11	6.19	2.04		99.5	17.99	6.15	2.14	

TABLE 8 SHOWING THE RELATION OF TIME OF PLANTING TO THE GLUTEN CONTENT OF WHEAT KERNELS

Sample No.	Early planting			Late planting		
	Total gluten	Total gliadin	Ash	Total gluten	Total gliadin	Ash
1	14.37	5.890	1.71	16.41	6.742	1.88
2-1	13.00	5.322	1.71	13.42	5.787	1.78
3	13.74	5.552	1.70	14.54	5.839	2.05
4-B	14.42	5.657	1.73	15.22	6.049	1.76
4-C	12.66	5.555	1.63	15.44	6.503	1.69
4-D	12.15	4.572	1.48	15.22	5.594	1.86
4-E	10.79	4.646	1.82	13.46	6.299	1.96
5-2	11.98	4.311	1.78	15.28	5.248	1.73
5-4	12.55	4.186	1.81	16.41	6.477	1.61
5-5	12.38	4.657	1.88	15.22	5.935	1.61
6	11.26	4.287	1.91	16.30	5.873	1.94
7	13.80	5.424	1.94	17.38	6.844	1.83
8	14.42	6.179	1.74	16.24	7.242	1.60
10-1	13.46	4.595	1.77	16.01	5.600	1.70
10-2	12.21	3.765	1.78	15.33	4.975	1.71
10-3	13.06	4.333	1.61	15.50	5.731	1.73
10-4	11.92	3.845	1.75	16.01	5.560	1.81
11	12.62	5.210	1.74	14.94	6.117	1.97
12-2	14.25	5.339	1.76	14.59	5.765	1.79
12-4	13.06	4.731	1.70	14.93	5.288	1.51
13	13.57	4.884	1.70	14.94	5.310	1.77
15	13.34	4.879	1.81	14.14	5.026	1.95
18	14.02	4.311	1.64	16.13	5.469	1.63
20	13.51	5.418	1.72	15.56	6.162	1.60
21	11.70	4.544	1.56	15.05	5.077	1.63
22	13.17	4.328	1.81	16.41	5.452	1.58
23	16.01	5.707	2.05	17.89	6.747	2.08
24	13.17	4.708	2.15	15.44	5.384	1.80
25	14.08	4.288	2.10	15.79	5.305	1.92
26	13.34	4.461	1.82	15.84	5.163	2.04
27	14.08	4.345	1.87	17.60	6.088	1.98
28	14.76	5.117	1.63	15.39	5.350	1.78
29	11.47	3.311	1.87	14.54	4.146	1.76
30	13.24	5.293	1.65	14.82	5.867	1.70
31	13.57	5.572	1.50	14.54	5.992	1.94
32	13.26	5.305	1.58	14.37	6.054	1.64
33	12.49	4.515	1.87	14.54	6.418	1.81
Av.	13.158	4.812	1.764	15.428	5.796	1.787

In this series of plantings, without a single exception, the late planted samples run higher in protein than those planted early, and the same holds true of their gliadin content.

## GENERAL DISCUSSION

Collecting all results for the season of 1908, 1909, and 1911, the following grand averages appear upon which to base conclusions:

Year	No. trials	First planting			Second planting		
		Per cent typical kernels	Per cent total protein	Per cent gliadin	Per cent typical kernels	Per cent total protein	Per cent gliadin
1908	17	78.5	12.75	3.84	84.3	13.45	4.26
1909	34	87.9	15.66	5.92	98.4	16.95	6.39
1911	37	.....	13.16	4.84	.....	15.43	5.80
Av.	88	83.2	13.85	4.87	91.3	15.28	5.48

These figures show quite definitely that late planting tends toward the production of a grain carrying a higher percentage of total protein and gliadin, as well as a higher percentage of typical kernels, than does early planting. Of the eighty-eight trials made, if we except those at Davis the first season when the two plantings were made at dates too close together for distinctive results, there is a great unanimity of results in favor of the late seeded grain in the respects indicated. Considered as a whole, the late plantings show an increase over the early plantings of 8.1 per cent of typical kernels, 1.43 per cent of protein and .61 per cent of gliadin. Of particular note are the four successive plantings made at Ceres in 1909, in which there is a regular increase in protein, gliadin, and typical kernels from the early to the latest plantings, and also the plantings at Davis in 1910-11 in which the same unanimity in respect to protein and gliadin is shown.

The results covering this season are so uniformly in favor of the late planting as developing the higher protein and gliadin content that the question seems to be very decisively answered.

#### THE EFFECT OF THE TIME OF HARVESTING UPON THE PROTEIN CONTENTS OF WHEATS

In most parts of the country from which is obtained the recognized high gluten wheat the grain is cut when in the hard dough stage by means of a self-binder and later threshed by means of a stationary threshing outfit. In California practically

all of the grain is left standing on the straw in the field until it is "dead ripe" and finally cut in that condition and threshed in a single operation by means of a combined harvester.

In the later method the grain often stands on the straw and is subjected to the action of the sun for several weeks after reaching the hard dough stage. This difference in the method of harvesting has led to the belief among many that the time of cutting might be one of the causes of the relatively low protein (gluten) content of Pacific Coast wheats.

With the idea of determining the effect of such standing in the field, a series of experiments was planned to ascertain what might be the influence of this practice.

For the purposes of this experiment several varieties of wheat whose kernels possessed distinctive characteristics as indicated in the preceding discussion were hand-separated into three groups as previously shown (page 69). These were drill-seeded under uniform soil and climatic condition at the several stations. One-half of each lot was harvested in the hard dough stage, in which condition it would normally be cut by a binder, and the other half was left standing on the straw in the field until it reached a condition suitable to be cut with a combined harvester. This was finally harvested and each portion subjected to analysis in addition to determining the percentage of typical kernels as indicated by the physical appearance of the kernels, using 1000 kernels as a basis.

Aside from the main question as to the effect of the time of cutting upon the quality of the grain, the data as presented in tabular form further furnishes a means for comparing the original used, with the progeny as in the previously discussed experiment (pages 69-74).

TABLE 9. SHOWING PHYSICAL AND CHEMICAL DEVIATION FROM TYPE IN EARLY AND LATE CUT WHEAT  
EXPERIMENT D 2, 1906

Lab. No.	Name	Per cent typical seed in original	Date of harvest	Per cent typical seed in progeny 1906	Number kernels in 10 grams	MODESTO			As analyzed			Calculated to dry basis		
						Moisture	Total nitrogen	Ash	Total nitrogen	Total protein	Ash	Total nitrogen	Total protein	Ash
709	Kubanka	100.0	June 27	97.7	252	10.98	2.33	13.24	1.99			2.62	14.91	2.24
710	Kubanka	100.0	July 31	88.7	204	10.95	2.10	11.96	1.90			2.36	13.40	2.13
711	Kubanka	0.0	June 27	89.4	216	10.87	2.31	13.16	1.98			2.59	14.75	2.22
712	Kubanka	0.0	July 31	69.7	195	10.90	1.98	11.24	1.94			2.22	12.60	2.18
713	Kubanka	0.0	June 27	96.9	195	10.95	2.23	12.68	1.89			2.50	14.20	2.12
714	Kubanka	0.0	July 31	100.0	185	10.63	2.37	13.48	1.90			2.65	15.10	2.13
715	Chul	100.0	June 12	94.1	219	11.96	1.82	10.36	2.00			2.07	11.76	2.27
716	Chul	100.0	July 31	83.4	.....	10.10	1.63	9.25	1.84			1.81	10.27	2.05
717	Chul	0.0	June 12	91.6	218	11.13	1.96	11.16	2.00			2.21	12.56	2.25
718	Chul	0.0	July 31	96.0	195	11.09	2.15	12.20	1.95			2.42	13.76	2.19
719	Chul	0.0	June 12	96.1	219	11.23	1.91	10.92	1.90			2.15	12.20	2.14
720	Chul	0.0	July 31	96.3	185	9.20	1.99	11.32	1.87			1.92	12.44	2.06
338A3	Turkey Red	100.0	June 27	98.0	314	11.28	2.08	11.80	2.08			2.34	13.32	2.34
338A4	Turkey Red	100.0	July 31	86.4	320	11.67	1.98	11.24	2.05			2.24	12.76	2.32
338B3	Turkey Red	0.0	June 27	91.1	309	11.19	1.98	11.24	2.02			2.23	13.64	2.27
338B4	Turkey Red	0.0	July 31	90.0	373	10.92	2.10	11.96	2.08			2.36	13.42	2.33
733	Turkey Red	100.0	June 27	99.3	306	10.33	2.16	12.28	2.02			2.40	13.63	2.25
734	Turkey Red	100.0	July 31	92.0	324	10.84	1.99	11.32	1.85			2.11	11.96	2.06
721	Frete	94.0	June 23	95.2	220	11.57	2.23	12.68	2.13			2.52	14.26	2.41
722	Frete	94.0	July 31	93.0	220	11.65	2.22	12.60	2.17			2.51	14.26	2.46
723	Koola	16.8	June 21	93.9	200	12.18	2.08	11.80	2.00			2.37	13.40	2.28
724	Koola	16.8	July 31	87.5	199	12.28	2.19	12.44	2.02			2.50	14.10	2.30
725	Kubanka	38.2	June 21	97.9	246	11.01	1.96	11.16	1.88			2.20	12.51	2.11
726	Kubanka	38.3	July 31	87.7	245	11.13	2.12	12.04	1.96			2.23	12.59	2.21
727	Maronani	43.2	July 14	93.1	171	11.50	2.15	12.20	1.98			2.43	13.71	2.28
728	Maronani	43.2	July 31	96.0	174	11.67	2.30	13.08	2.05			2.60	14.72	2.30
729	Red Winter	51.0	June 21	91.5	293	11.32	2.12	12.04	2.05			2.39	13.55	2.31
730	Red Winter	51.0	July 31	88.0	276	11.43	1.88	10.68	2.06			2.12	12.04	2.32
731	.....	78.7	June 27	93.5	263	11.21	2.10	11.96	2.03			2.37	13.40	2.28
732	.....	78.7	July 31	97.0	297	11.04	2.29	13.00	2.06			2.56	14.51	2.30

TABLE 9. SHOWING PHYSICAL AND CHEMICAL DEVIATION FROM TYPE IN EARLY AND LATE CUT WHEAT—(Concluded)

Lab. No.	Name	Per cent typical seed in 1906	Date harvest	Per cent typical kernels in original	YUBA CITY			As analyzed			Calculated to dry basis		
					Number kernels per bushel	Moisture	Total nitrogen	Total protein	Ash		Total nitrogen	Total protein	Ash
645	Kubanka	100.0	July 2	55.6	217	10.11	1.92	10.92	2.04		2.14	12.12	2.26
646	Kubanka	100.0	Aug. 13	52.1	199	10.26	1.83	10.36	2.08		2.03	11.56	2.32
648	Kubanka	0.0	July 2	50.8	197	10.38	1.82	10.36	1.98		2.03	11.56	2.21
649	Kubanka	0.0	Aug. 13	44.1	187	10.31	1.78	10.11	1.95		1.99	11.32	2.19
651	Kubanka	0.0	July 2	63.5	205	10.30	1.89	10.76	2.02		2.11	12.04	2.25
652	Kubanka	0.0	Aug. 13	61.7	200	10.28	1.88	10.68	2.06		2.09	11.88	2.29
654	Chul	100.0	June 30	69.4	203	10.50	1.81	10.27	1.99		2.03	11.56	2.22
655	Chul	100.0	Aug. 13	63.0	195	10.67	1.81	10.27	1.98		2.02	11.48	2.22
657	Chul	0.0	June 30	76.8	221	10.31	1.94	11.00	2.00		2.17	12.36	2.12
658	Chul	0.0	Aug. 13	65.1	218	10.65	1.78	10.11	1.88		1.99	11.32	2.10
660	Chul	0.0	June 30	52.0	.....	12.49	1.87	10.64	2.00		2.12	12.04	2.28
661	Chul	0.0	Aug. 13	62.3	.....	12.43	1.82	10.36	1.94		2.06	11.72	2.22
338A	Turkey Red	100.0	July 7	65.3	300	10.41	1.77	10.03	1.87		1.98	11.25	2.09
338A1	Turkey Red	100.0	Aug. 13	69.6	298	10.40	1.78	10.11	1.91		1.98	11.25	2.13
338B	Turkey Red	0.0	July 7	67.3	300	10.15	1.77	10.03	1.97		1.97	11.20	2.19
338B1	Turkey Red	0.0	Aug. 13	50.1	302	10.41	1.78	11.11	1.94		1.98	11.52	2.17
663	Fretes	50.8	June 30	100.0	217	10.10	2.02	11.48	2.10		2.25	12.80	2.34
664	Fretes	50.8	Aug. 13	100.0	213	10.37	2.06	11.72	2.05		2.30	12.68	2.29
666	Kharkov	37.5	July 10	93.1	301	10.47	2.13	12.12	2.15		2.38	13.68	2.40
667	Kharkov	37.5	Aug. 13	88.7	278	10.46	2.08	11.80	2.08		2.32	13.32	2.23
669	Weissenberg	15.8	July 10	87.7	304	10.04	2.09	11.88	2.18		2.32	13.32	2.42
670	Weissenberg	15.8	Aug. 13	79.1	300	10.10	2.95	11.04	2.17		2.16	13.26	2.41
672	Kahla	75.2	July 2	53.0	201	10.04	2.07	11.76	2.19		2.30	12.68	2.43
673	Kahla	75.2	Aug. 13	79.1	197	10.09	2.62	12.84	2.22		2.52	14.31	2.47
675	Gharnovka	.....	July 5	15.3	240	11.28	1.55	8.80	2.02		1.75	9.94	2.78
676	Gharnovka	.....	Aug. 13	9.0	235	11.09	1.53	8.70	2.01		1.72	9.77	2.26
678	Kubanka	23.7	July 2	63.4	215	10.31	1.91	10.84	2.05		2.13	12.12	2.29
679	Kubanka	23.7	Aug. 13	71.8	180	10.57	2.13	12.12	2.20		2.38	13.68	2.46
681	Beloglina	.....	July 5	66.1	305	10.50	1.66	9.41	1.98		1.85	9.43	2.21
682	Beloglina	.....	Aug. 13	59.9	299	10.49	1.81	10.27	2.00		2.02	10.28	2.23
684	Velvet Don	93.8	July 2	79.5	217	10.05	2.02	11.98	1.95		2.25	13.74	2.17
	Velvet Don	93.8	Aug. 13	78.2	224	10.04	2.03	11.56	1.89		2.26	13.73	2.10

*Discussion of 1906 Results.*—In these results the same thing is noticeable as in the former series, viz., that the samples grown at Modesto as a whole maintained both a higher percentage of typical kernels and a higher protein content than those grown at Yuba City, the comparison by averages being as follows:

	Modesto	Yuba City
Average per cent typical kernels .....	89.70	65.2
Average per cent protein .....	11.87	10.5

It is particularly noticeable that, outside of the instances in which the originals carried 100 per cent of typical kernels, there was in this season a practically universal marked increase in the number of typical kernels in the progeny; that in the case of sample no. 714, which carried no typical kernels, the original seed being 100 per cent starchy, the second cutting produced 100 per cent of typical kernels and the first cutting (no. 713) 96.9 per cent of such kernels. Almost as good a showing also is made by samples nos. 719 and 720, which produced respectively 96.1 and 96.3 typical kernels from entirely starchy kernels.

It is further shown that in the case of the early cutting twenty-one out of the thirty-one trials showed an increase in the percentage of typical kernels in the progeny over the original, the relation in those cases in which increase occurred being 42.5 per cent in the original and 83.19 per cent in the progeny, while in the eight cases in which decrease occurred the averages were 96.1 per cent typical kernels in the originals against 75.3 per cent in the progeny, which is entirely confirmatory of the results presented in Table 2.

Looking more particularly at the specific question involved in this experiment, viz., the effect of the time of cutting upon the percentage of typical kernels and the protein content, it is found that the following averages hold:

MODESTO			
	Number of trials	Per cent typical kernels	Per cent protein
Early cutting .....	15	94.6	13.38
Late cutting .....	15	90.1	13.19



## YUBA CITY

	Number of trials	Per cent typical kernels	Per cent protein
Early cutting .....	16	66.1	11.98
Late cutting .....	16	64.6	12.05

These average figures show, in the case of the Yuba City lot, an apparent contradiction in that the early cutting with 66.1 per cent typical kernels showed 11.98 per cent protein, while the late cutting with but 64.6 typical kernels showed 12.05 per cent protein. When it is remembered, however, that the difference in the two cuttings is represented by but a single kernel in one hundred and that the average protein content is but seven-hundredths of 1 per cent, it will be seen that the discrepancy is only apparent, and well within the limits of error in either count or analysis.

As a whole, it is shown that out of the total of 31 cases 11 showed an increase of typical kernels in the late cut grain and 14 showed an increased protein content, with the following relative averages:

	Number of trials	Per cent typical kernels	Per cent protein
Early cuttings .....	31	79.89	12.66
Late cuttings .....	31	76.9	12.60

The results for 1906 do not show that there is any marked difference to be attributed to allowing the grain to become thoroughly ripe on the straw, as is generally done in California.

## EXPERIMENTS OF 1907

The experiments as to the effect of the time of cutting wheat upon its protein content were continued in the manner described above in the season of 1907 at Modesto, Yuba City, and Tulare, with the following results:

TABLE 10. SHOWING PHYSICAL AND CHEMICAL DEVIATION FROM TYPE IN EARLY AND LATE HARVESTED WHEATS  
MODESTO, 1907

Lab. No.	Name	Per cent typical seed in original	Date of harvest	Per cent typical seed in progeny	Number kernels in 10 grams	As analyzed					Calculated to dry basis			
						Moisture	Total nitrogen	Total protein	Gladiin	Ash	Total nitrogen	Total protein	Gladiin	Ash
924A	Chul	100.0	June 10	49.0	242	11.89	1.53	8.71	2.50	1.88	1.74	9.88	2.81	2.13
924B	Chul	100.0	June 20	55.0	221	12.02	1.47	8.38	2.66	1.94	1.75	9.96	3.02	2.20
925A	Chul	0.0	June 10	42.0	220	11.93	1.42	8.05	1.49	1.83	1.60	9.14	1.69	2.07
925B	Chul	0.0	July 20	42.0	215	11.86	1.51	8.57	2.41	1.82	1.71	9.72	2.73	2.06
926A	Chul	0.0	June 10	60.0	227	11.61	1.71	9.74	3.19	2.04	1.95	11.02	3.61	2.29
926B	Chul	0.0	July 20	53.0	232	11.70	1.64	9.33	2.90	1.95	1.86	10.56	3.28	2.21
927A	Kubanka	100.0	June 21	48.0	222	11.95	1.71	9.74	3.00	1.76	1.94	11.06	3.41	1.99
927B	Kubanka	100.0	July 20	35.0	241	11.42	1.67	9.50	2.30	1.74	1.88	10.73	2.59	1.96
928A	Kubanka	0.0	June 21	49.0	249	11.50	1.70	9.66	1.75	1.74	1.92	10.92	1.97	1.97
928B	Kubanka	0.0	July 20	54.0	273	11.73	1.59	9.04	3.16	1.94	1.80	10.24	3.46	2.19
929A	Kubanka	0.0	June 21	36.0	242	11.52	1.69	9.61	2.98	1.83	1.91	10.87	3.37	2.07
929B	Kubanka	0.0	July 20	35.0	241	11.72	1.62	9.22	3.00	1.83	1.84	10.45	3.39	2.07
930A	Fretes	83.3	June 21	63.0	235	11.99	1.77	10.06	4.11	1.91	2.01	11.43	4.89	2.27
930B	Fretes	83.3	July 20	63.0	230	12.09	1.85	10.51	3.70	1.93	1.93	11.95	4.13	2.15
931A	Erivan	80.9	June 21	73.0	365	11.56	1.61	9.21	2.93	1.95	1.83	10.42	3.31	2.20
931B	Erivan	80.9	July 20	67.0	363	11.50	1.57	8.93	2.51	2.02	1.77	10.09	2.83	2.28
932A	Gharnovka	21.4	June 21	37.0	266	12.07	1.60	9.07	2.98	1.68	1.81	10.32	3.39	1.89
932B	Gharnovka	21.4	July 20	.....	267	11.66	1.74	9.93	3.20	1.73	1.98	11.34	3.62	1.95
933A	Koola	87.0	June 21	52.0	255	11.88	1.81	10.28	3.24	1.91	2.05	11.66	3.67	2.16
933B	Koola	78.0	July 20	45.0	275	12.02	1.48	8.43	3.22	1.84	1.69	9.58	3.66	2.09
934A	Kubanka	78.0	June 21	.....	225	11.75	1.65	9.34	3.61	1.74	1.87	10.59	4.09	1.97
934B	Kubanka	87.0	July 20	.....	218	11.51	1.60	9.09	3.00	1.81	1.81	10.27	3.39	2.04
935A	Velvet Don	44.1	June 21	51.0	280	11.78	1.81	10.28	2.95	1.78	2.05	11.65	3.34	2.02
935B	Velvet Don	44.1	July 20	43.0	273	11.61	1.57	8.93	2.96	1.74	1.78	10.11	3.34	1.96

TABLE 10.—SHOWING PHYSICAL AND CHEMICAL DEVIATION FROM TYPE IN EARLY AND LATE HARVESTED WHEATS—(Continued)

YUBA CITY, 1907

Lab. No.	Name	Per cent typical seed in original	Date of harvest	Per cent typical progeny	Number kernels in 10 grams	As analyzed				Calculated to dry basis				
						Moisture	Total nitrogen	Total protein	Ash	Total nitrogen	Total protein	Gladiin	Ash	
847	Kubanka	100.0	July 2	48.6	.....	13.24	1.69	9.58	3.37	1.71	1.95	11.03	3.88	1.97
848	Kubanka	100.0	July 24	49.5	.....	12.80	1.73	9.82	2.83	2.02	1.99	11.26	3.31	2.32
850	Kubanka	0.0	July 2	48.3	203	13.08	1.61	9.20	1.90	1.67	1.87	10.58	2.17	1.92
851	Kubanka	0.0	July 24	42.3	210	13.09	1.76	10.00	3.01	1.62	2.02	11.51	3.55	1.90
853	Kubanka	0.0	July 2	38.9	224	13.50	1.60	9.07	2.79	1.98	1.60	9.07	3.23	2.28
854	Kubanka	0.0	July 24	50.3	219	12.99	1.67	9.47	2.99	1.77	1.77	10.07	3.43	2.02
856	Chul	100.0	June 29	58.1	.....	12.98	1.49	8.47	3.49	2.01	1.71	9.74	.....	2.31
856	Chul	100.0	July 12	55.8	192	14.08	1.24	7.04	2.83	1.63	1.45	8.19	3.29	1.89
857	Chul	0.0	June 29	87.7	200	13.88	1.19	6.73	2.97	1.49	1.37	7.81	3.45	2.15
858	Chul	0.0	July 12	68.0	200	13.71	1.28	7.29	2.78	1.74	1.49	8.44	3.23	2.02
861	Kharkov	93.1	Aug. 7	83.9	310	11.64	2.07	11.75	3.16	2.07	2.19	12.44	3.66	2.40
860	Kharkov	93.3	Sept. 13	86.1	276	11.18	.....	.....	.....	.....	.....	12.70	3.63	2.28
862	Weissenberg	87.7	July 7	89.1	270	13.67	2.44	13.87	4.36	2.06	2.82	16.06	5.06	2.38
863	Weissenberg	87.7	Sept. 6	99.3	278	12.79	3.06	17.53	4.23	2.17	3.52	20.10	4.75	2.48
866	Fretes	100.0	July 2	83.5	225	12.85	1.73	9.82	3.17	2.12	1.98	11.27	3.72	2.43
867	Fretes	100.0	Sept. 6	63.1	147	12.82	1.70	9.64	3.39	1.97	1.96	11.05	3.88	2.26
868	Kubanka	63.4	July 8	89.2	184	13.36	3.57	20.48	4.05	1.95	4.14	23.64	4.68	2.22
869	Kubanka	63.4	Sept. 5	88.3	179	.....	.....	.....	.....	.....	.....	.....	.....	.....
870	Velvet Don	79.5	July 2	90.1	239	12.84	2.14	12.17	4.12	2.27	2.46	13.96	4.75	2.61
871	Velvet Don	79.5	Aug. 6	96.1	216	13.17	2.09	11.94	4.53	2.22	2.43	15.76	5.21	2.56
872	Gharnovka	6.5	July 2	66.9	210	12.06	1.91	10.84	3.08	1.74	2.12	12.05	3.50	.....
873	Gharnovka	6.5	Aug. 6	76.5	206	13.70	1.91	10.83	2.68	2.14	2.20	12.46	3.12	.....
874	Beloglina	59.9	July 23	77.4	314	12.72	1.83	10.40	3.42	2.21	2.09	11.91	3.91	2.54
876	Beloglina	59.9	Sept. 7	91.9	356	11.82	2.06	11.71	1.95	2.25	2.33	13.27	2.21	2.55
877	Currell	.....	July 22	87.9	323	13.25	2.07	11.73	4.34	1.84	2.38	13.52	4.00	2.12
879	Currell	.....	Sept. 13	78.6	351	13.11	2.08	11.78	4.39	1.90	2.39	13.55	5.05	2.02

TABLE 10.—SHOWING PHYSICAL AND CHEMICAL DEVIATION FROM TYPE IN EARLY AND LATE HARVESTED WHEATS—(Concluded)

Lab. No.	Name	Per cent typical seed in original	Date of harvest	Per cent typical seed in progeny	Number kernels in 10 grams	As analyzed					Calculated to dry basis			
						Moisture	nitrogen	Total protein	Gliadin	Ash	Total nitrogen	Total protein	Gliadin	Ash
892	Beloglina	66.1	June 21	59.4	271	13.21	1.67	9.51	3.33	2.04	1.93	10.96	3.83	2.35
893	Beloglina	66.1	July 29	57.7	278	13.71	1.60	9.14	3.19	1.73	1.86	10.96	3.69	2.00
894	Velvet Don	79.8	June 6	93.5	249	13.31	1.48	8.10	2.63	1.97	1.65	9.34	3.04	2.27
895	Velvet Don	79.8	July 29	84.8	226	12.91	1.87	10.59	3.77	1.86	2.15	12.17	4.33	2.26
899	Gharnovka	9.0	June 11	44.1	303	12.07	1.53	8.75	3.56	1.97	1.87	10.71	4.04	2.23
900	Gharnovka	9.0	July 29	46.8	232	12.97	1.76	10.00	3.12	2.07	2.12	12.06	3.58	2.37
901	Turkey Red	50.1	June 27	85.1	310	13.49	2.04	11.58	4.10	2.16	2.36	13.39	4.85	2.49
902	Turkey Red	50.1	July 29	66.9	283	14.14	1.76	10.01	3.18	1.82	2.06	11.71	3.70	2.12
904	Fretea	100.0	June 3	0.0	220	13.87	1.61	9.17	3.60	1.86	1.87	10.60	4.19	2.16
905	Fretea	100.0	July 29	32.7	229	13.54	1.23	7.00	3.41	1.70	1.42	8.10	4.06	1.95

The averages for the three localities are shown in the following small table:

TABLE SHOWING THE AVERAGE RESULTS FROM EARLY AND LATE CUT WHEATS  
AT THREE STATION IN 1907

	Number of trials	Early cut		Late cut	
		Per cent typical kernels	Per cent protein	Per cent typical kernels	Per cent protein
Modesto	12	52.3	9.92	49.2	10.40
Yuba City	13	73.0	11.62	73.3	12.19
Tulare	5	56.4	11.00	57.8	10.92
Grand av.	30	62.6	10.80	61.9	11.24

From a total of 30 cases, 12 showed either an equal or larger per cent of typical kernels in the late cutting than in the early cutting, a number altogether too large to indicate that the lateness of cutting had any material influence in this direction, and this is further shown from the fact that the general average shows 62.6 per cent of typical kernels in the early cut lots and 61.9 per cent in the late cut lots.

Again this is indicated in the protein content, for out of the 30 cases 15 show a larger percentage in the late cut grain than in the early cut lots, and the general average in protein content is essentially the same in the two lots, viz., 10.80 and 11.24 per cent respectively.

For the two seasons the record stands as follows:

	Number of trials	Early cut		Late cut	
		Per cent typical kernels	Per cent protein	Per cent typical kernels	Per cent protein
Modesto	27	77.68	11.8	73.7	11.95
Yuba City	29	69.1	11.96	68.5	11.20
Tulare	5	56.4	11.00	57.8	10.92
Average	61	71.1	11.80	69.9	11.51

These figures seem to show quite clearly that to allow the grain to stand on the straw until in the proper condition for handling with the combined harvester does not in any manner militate against its quality either in physical appearance or protein content, and should set at rest any further discussion upon this point.

## EFFECT OF SUNSHINE ON THE COMPOSITION OF THE WHEAT KERNEL

It has long been a matter of common knowledge that the composition of plants varies greatly under different conditions and in different localities. The exact environmental factor or factors which cause this variation in composition has been the cause of much discussion and investigation.

It has already been remarked above that in 1882 Richardson observed that wheat grown in Colorado had by reputation a much higher gluten content than wheat grown from the same seed in Oregon. Not only was there a marked difference in the composition of the wheat grown in the two states, but he found that the wheat grown in Colorado had a higher gluten content than the original seed, while that grown in Oregon had a lower gluten content than the seed from which it was produced. From these observations Richardson and Blount concluded that the soil was the modifying factor.

Wiley, however, draws the conclusion that the difference in gluten content is due to climatic conditions.

Lawes and Gilbert in an elaborate series of experiments have shown that the use of manures and fertilizers have very little influence on the composition of the wheat kernel. On the other hand, they found a wide variation in composition in different seasons.

Deherain in France also observed that a difference in seasonal conditions, especially during the ripening period, had a marked influence on the gluten content of wheat.

Similar observations have been noted by Thatcher of Washington, and others.

From these numerous observations and experiments it has come to be generally conceded that the gluten content is influenced mostly, if not wholly, by the climate.

Wheat grown in the coast states is, as a class, much lower in gluten content than wheat grown in the central west, or the northwestern states. Even when seed of a high gluten content is introduced, a product, as Richardson observed, considerably inferior is the result.

Climate includes a large number of factors. The specific climatic factor which is the cause of this variation has recently been the subject of much study and investigation.

The formation of organic compounds in the plant, such as starch and gluten, is a physiological process. The maximum development of these compounds is necessarily dependent upon favorable conditions. Starch is formed under the influence of sunlight. The formation of nitrogenous compounds requires not only an adequate supply of nitrogen, but also a supply of carbon compounds in the proper form and under the proper conditions. Just what these conditions are is not definitely known.

In general, investigators have concluded that the large amount of sunshine prevalent in the coast states during the period in which the seed develops works directly in favor of the formation of large amounts of starch. In other words, that the gluten content is low only by reason of the formation of proportionally larger amounts of starch. Were this the case, the exclusion of portions of the sunlight should tend to increase the percentage of gluten. Theoretically the gluten would increase inversely with the amount of sunlight which the plants received.

In order to determine whether or not sunlight is a prominent factor a series of experiments were planned in which portions of the natural sunlight were excluded from the growing plants.

Duplicate experiments were conducted during the seasons of 1908 and 1909 at the Tulare Sub-station and at the University Farm at Davis.

*Original Seed.*—At the Tulare Station in 1908 the originals used were as follows:

No. 864/07	Weissenberg
No. 879/07	Currell
No. 892/07	Beloglina
No. 894/07	Velvet Don (starch grains)
No. 901/07	Turkey Red (gluten grains)
No. 899/07	Yellow Gharnovka (gluten grains)

At Davis the following varieties were used:

No. 868/07	Kubanka
No. 847	Kubanka
No. 870A	Velvet Don (gluten grains)

The originals showed the following composition:

#### AT TULARE

Lab. No.	Name	Per cent typical kernels	Kernels in 10 grams	Per cent protein	Per cent gliadin	Per cent ash
864	Weissenberg	95.9	325	15.25	3.63	2.66
879	Currell	87.6	351	13.55	5.06	2.11
892	Beloglina	59.4	271	10.96	3.84	2.33
894	Velvet Don	0.0	230	8.79	2.90	2.00
901	Turkey Red	100.0	278	13.41	5.28	1.99
899	Gharnovka	100.0	280	12.20	4.26	2.02

#### AT DAVIS

Lab. No.	Name	Per cent typical kernels	Kernels in 10 grams	Per cent protein	Per cent gliadin	Per cent ash
868	Kubanka	89.2	184	23.64	4.67	2.22
847	Kubanka	48.6	.....	11.03	3.88	1.97
870-G	Velvet Don	90.1	239	17.38	4.80	2.05

Two rows of each of these varieties were drilled parallel into a bed 24 feet long. In the spring after the grain began to make an upright growth a series of lath screens was constructed and placed across the plat so as to shut off different proportions of the direct sunshine. The plat was thus divided into sections. At Tulare sectional screens were arranged so that they excluded three quarters, one half, and one quarter of the sunshine respectively. One quarter was left unshaded, and this received the full sunshine. In this manner one fourth of each variety received the same amount of sunshine. At Davis a similar arrangement was used, except that the amount of sunshine admitted was one third, one half, two thirds, and full. These screens were left in place until the grain was matured and harvested. Each portion of each variety thus treated was harvested separately and taken to the laboratory for analysis. The results are stated below:



TABLE 12.—SHOWING EFFECT OF VARIOUS AMOUNTS OF SUNSHINE ON THE GLUTEN CONTENT OF WHEAT

TULARE, 1908								
	One-quarter sunshine		One-half sunshine		Three-quarters sunshine		Full sunshine	
	Total gluten	Gliadin	Total gluten	Gliadin	Total gluten	Gliadin	Total gluten	Gliadin
864	17.55	8.000	18.00	8.065	.....	.....	20.73	9.599
894	12.10	4.930	12.72	5.350	.....	.....	13.57	4.856
899	12.66	5.333	12.61	5.311	.....	.....	13.63	5.515
901	15.33	6.602	15.45	6.986	.....	.....	16.13	6.545
879	18.17	8.000	18.63	8.406	.....	.....	18.34	8.463
892	15.39	6.674	16.58	7.157	.....	.....	17.04	7.554
Av.	15.19	6.589	15.665	6.879	.....	.....	16.57	7.088
Gliadin ratio	43.40		43.91		.....		42.77	

DAVIS, 1908								
	One-third sunshine		One-half sunshine		Two-thirds sunshine		Full sunshine	
	Total gluten	Gliadin	Total gluten	Gliadin	Total gluten	Gliadin	Total gluten	Gliadin
868	13.19	4.17	13.94	4.17	14.10	4.42	13.79	4.28
847	12.69	4.46	12.37	4.09	13.40	3.50	12.47	4.00
870G	14.30	4.71	.....	.....	13.53	4.21	13.17	4.52
Av.	13.39	4.445	13.15	4.13	13.67	4.034	13.14	4.266
Gliadin ratio	33.19		31.40		29.57		32.46	

## EXPERIMENTS OF 1909

During the season of 1909 the same experiment was conducted at the Tulare and Davis Stations, with the exception that the following varieties of wheat were used:

Gluten grains from Richi 1044A;  
 Gluten grains from Kubanka 1045A;  
 Starch grains from Kubanka 1045A;  
 Gluten grains from Turkey 1046A;  
 Gluten grains from Beloglina 1047A;  
 Starch grains from Beloglina 1047A.

Determinations made upon these previous to seeding showed as follows:

No.	Typical kernels	Kernels in 10 grams	Protein	Gliadin	Ash
1044A-G	100.0	189	10.42	3.17	1.90
1045A-G	100.0	197	13.09	4.32	1.74
1045A-S	0.0	220	9.37	2.59	1.88
1046A-G	100.0	389	15.43	4.90	2.21
1047A-G	100.0	372	16.11	5.33	2.25
1047A-S	0.0	329	7.37	2.00	1.77

At Davis the originals and their analyses were as follows:

No.	Name	Typical kernels	Kernels in 10 grams	Protein	Gliadin	Ash
870G	Velvet Don	100.0	227	15.92	4.27	2.02
870S	Velvet Don	0.0	239	13.96	4.73	2.61
1045A-G	Kubanka	100.0	197	13.09	4.32	1.74
1045A-S	Kubanka	0.0	220	9.37	2.59	1.88
1041A-G	Kubanka	100.0	177	13.05	4.12	2.02
1041A-S	Kubanka	0.0	194	10.56	2.99	2.01

The gluten and gliadin content of the wheat samples from the experiments of 1909 are reported in Table 13.

TABLE 13.—SHOWING EFFECT OF VARIOUS AMOUNTS OF SUNSHINE ON THE PROTEIN CONTENT OF WHEAT

TULARE, 1909

	One-quarter sunshine		One-half sunshine		Three-quarters sunshine		Full sunshine	
	Total protein	Gliadin	Total protein	Gliadin	Total protein	Gliadin	Total protein	Gliadin
1046G	10.62	3.516	7.38	5.043	.....	.....	14.77	5.651
1047AS	10.96	3.998	12.32	4.413	15.56	5.577	13.18	4.084
1047AS	.....	.....	.....	.....	.....	.....	13.74	4.737
1045AS	11.64	4.100	12.04	4.271	13.86	3.783	13.35	3.839
1045AG	12.38	3.902	13.25	4.674	11.30	4.312	13.91	4.981
1044AG	10.90	3.271	11.41	3.436	14.99	4.385	11.59	3.720
Av.	11.30	3.757	11.27	4.368	13.92	4.514	13.42	4.502
Gliadin ratio	33.42		38.74		32.42		33.54	

DAVIS, 1909

	One-third sunshine		One-half sunshine		Two-thirds sunshine		Full sunshine	
	Total protein	Gliadin	Total protein	Gliadin	Total protein	Gliadin	Total protein	Gliadin
1041AS	15.34	4.845	16.24	4.850	16.41	5.151	16.02	4.970
1045AG	14.71	5.186	14.37	4.748	15.50	4.055	14.48	4.635
1045AS	14.71	5.680	* .....	.....	15.56	6.248	14.25	5.850
870AG	16.53	5.341	* .....	.....	15.62	4.867	15.66	5.214
870AS	15.79	6.816	14.06	3.770	15.50	6.418	15.50	6.645
1041AG	16.36	6.832	16.70	6.475	17.09	6.929	16.30	6.418
Av.	15.57	5.773	15.34	4.960	15.94	5.611	15.37	5.622
Gliadin ratio	36.82		32.33		35.20		36.57	

\* There was not enough material for a re-analysis of the sample.

*Discussion.*—If the proportion of protein to starch, or the percentage of nitrogen, increased inversely as the amount of sunshine the plants received we would expect the percentage of protein to be highest in the plants receiving only one quarter of the total sunshine and to decrease gradually to full sunshine. This, however, does not seem to be the case. Taking the average of the six samples grown at the Tulare station in 1908, we find just the opposite result. The protein content increases with the sunshine quite uniformly. The wheat from the plants receiving one-half sunshine contain .47 per cent more protein than the wheat from those receiving one-quarter sunshine. The wheat receiving full sunshine contained .91 per cent more than those receiving only one-half sunshine. Unfortunately the samples grown under three-quarters sunshine were lost, so that we are unable to say whether or not the protein content here would have been above or below that of full sunshine.

The analysis of the individual samples in this set show that only one departs markedly from the average. In the variety no. 879 we find that the wheat grown under one-half sunshine had a higher protein content by .29 per cent than did that grown under full sunshine, while that grown under one-quarter sunshine is the lowest of the three by .17 per cent. In sample no. 899 we find that one-quarter and one-half sunshine gave nearly the same results, with a slight difference in favor of the former.

The general trend of the results from 1909 experiments is much the same. The average of the samples grown under one-quarter and one-half sunshine are about equal, while the protein content of those grown under three quarters and full sunshine are much higher. If we exclude sample no. 1046G grown under one-half sunshine, which is unusually low, the average for one-half sunshine is 12.25 per cent. This gives, then, quite a gradual increase in protein content from one quarter to three quarters sunshine, while the full sunshine samples average .5 per cent lower than those receiving only three-quarters sunshine.

The results from the experiments at Davis are slightly more erratic, but the general trend is the same as that obtained at Tulare during the season of 1909. The samples under one third sunshine average .24 per cent more protein than did those under

one-half sunshine. The samples under two-thirds sunshine averaged .52 per cent more protein than those under one-half sunshine, and .28 per cent more than those under one-third sunshine. There was, then, a drop in the full sunshine sample to about the same protein content as those receiving one-half sunshine.

The results for 1909 at Davis came in the same order as those of 1908. The samples under one-third sunshine average higher in protein than those under one-half sunshine by .23 per cent. The samples under two-thirds sunshine averaged higher than those under one-half by .6 per cent, and than those under one-third sunshine by .37 per cent, while the average of the samples grown under full sunshine is nearly the same as of those grown under one-half sunshine.

These results certainly show that the protein content does not vary inversely with the amount of sunshine which the plants receive. On the other hand, the experiments at Davis and those of the year 1909 at Tulare tend to show that there is a happy medium under which the maximum amount of protein is stored. This optimum condition seems to be at a point somewhat below the normal sunshine. If, however, the amount of sunshine falls below that medium then again there is a decrease in the amount of protein stored. Just why this should be the case is still a matter of conjecture. It is quite probable, however, that it is due to a disturbed condition of the physiological functions within the plant brought about by the abnormally low sunshine. This fact can only be determined by a closer study of the formation and transformation of the various compounds in the plant.

The gliadin content of the samples in this experiment seems to bear even less relation to the sunshine than did the total protein. In fact, there was no regularity whatever in the results those obtained at Tulare being just opposite from those obtained at Davis.

A comparison of the two years at the two stations shows that there was a marked difference in the protein content of the samples in different seasons. At Davis we find that the samples averaged a higher percentage of gluten in 1909 than in 1908 by over 2 per cent.

Comparing the weather reports at Davis for the months of April, May, and June, we find that this period during 1909 was much drier and somewhat warmer than the corresponding months the year before. As this is the time when the greatest development of the kernel takes place, it is not at all unlikely that these climatic differences may account to some extent for the differences in composition of the grain. The moister and cooler condition of 1908 may have prolonged the developing and ripening period, thus favoring the storing of a larger amount of starch. Other seasonal differences not recorded have doubtless also contributed their share.

The difference in protein content for the two seasons at Tulare is just as marked as at Davis, but the order is reversed. No weather reports from the immediate vicinity were available, so we can make no comparison with the Davis conditions.

Another rather striking seasonal difference brought out by the table is the percentage of gliadin to total protein. The years that the percentage of total protein is high, the percentage of gliadin to total gluten is also high. In fact, the difference in the gliadin content of the wheat samples for the two years is proportionally greater than the difference in total protein. This would lead us to believe that the gliadin is affected more by the season than the other protein compounds. We find, however, little or no relation between the gliadin content and the sunshine received.

In conclusion, then, it is safe to say that while sunshine does exert some influence upon the composition of the wheat grain, there are other climatic factors which also exert very marked influences in this direction. We find that the protein does not increase inversely as the sunshine, but that there is an optimum condition under which the greatest development of protein takes place. This optimum of sunshine is somewhat less than normal in the valleys of this state. Other things being equal, too little sunshine lowers the protein content to just as great an extent as too much sunshine. This condition is probably due to the fact that a certain amount of sunshine is necessary in order that the normal physiological functions of the plant may take place. When the amount of sunshine is reduced to one quarter or one

half of the normal it is quite likely that the plants do not receive enough sunshine to allow even the maximum nitrogen metabolism to take place.

The fact that there is a greater difference in the percentage of protein for different seasons than there is in the same season under various amounts of sunshine certainly tends to show that there are factors other than sunshine, which play just as important a part in determining the composition of the grain.

Certain experimenters have stated that the exposure of grain to the action of strong sunlight after it had become ripe had a tendency not only to bleach the kernels, forming the so-called "yellow berry," but also to lower the protein content. Attention was called to this by Lyon and Keyser based upon some trials made at the Nebraska station. On account of the fact that the sunshine in the main grain-growing regions of California is very intense during the ripening and harvesting period, and that the grain frequently stands for several weeks on the straw in the field, an attempt was made in 1906 at Yuba City to ascertain if this effect held under the conditions which obtain in this state. In this experiment several varieties of wheat were selected in which differences in the physical appearances of the grain could be easily followed owing to the color of the typical and changed kernels. The percentage of typical kernels in the original was determined, using 5000 kernels in each case as a basis, and these were seeded in plats under like conditions in the field. At the maturity of the grain three fourths of the grain was cut while in the hard dough stage, a few bundles being shocked and left in the field, while an equal number were protected from the direct action of the sunlight. The remainder of the grain was left on the standing straw from July 5 to August 13, when it was harvested and all three lots were threshed at the same time. The experiment was repeated again in 1907. A determination was made of the percentage of typical kernels in each lot, and of the percent of protein. The results follow:

TABLE 14. SHOWING THE PHYSICAL AND CHEMICAL COMPOSITION OF WHEAT KERNELS EXPOSED AND PROTECTED FROM DIRECT SUNLIGHT  
SEASON OF 1906

Lab. No.	Name	Treatment	Date of harvest	Typical kernels Per cent	As analyzed				Calculated to dry matter		
					Moisture	Total nitrogen	Total protein	Ash	Total nitrogen	Total protein	Ash
681	Beloglina	Cut and left exposed	July 5	62.1	11.03	1.78	10.11	2.03	2.00	11.40	2.28
695	Beloglina	Cut and protected	July 5	66.1	10.63	1.94	11.02	2.07	2.19	12.48	2.34
682	Beloglina	Left on straw in field	Aug. 13	59.9	10.49	1.81	10.27	2.00	2.02	11.51	2.24
696	Gharnovka	Cut and left exposed	July 5	6.5	11.28	1.55	2.80	2.02	1.74	9.92	2.27
675	Gharnovka	Cut and protected	July 5	15.3	11.04	1.48	8.38	1.90	1.67	9.52	2.13
676	Gharnovka	Left on straw in field	Aug. 13	9.0	11.09	1.56	8.89	2.01	1.75	9.98	2.26

TABLE 14.—SHOWING THE PHYSICAL AND CHEMICAL COMPOSITION OF WHEAT KERNELS EXPOSED AND PROTECTED FROM DIRECT SUNLIGHT—(Continued)

Lab. No.	Name	Treatment	Date of harvest	Typical kernels Per cent	As analyzed			Calculated to dry matter		
					Moisture	Total nitrogen	Total protein	Ash	Total nitrogen	Total protein
861	Kharkov	Cut and left exposed	Aug. 7	83.9	13.64	1.89	10.75	2.07	2.18	12.44
859	Kharkov	Cut and protected	Aug. 6	92.4	12.03	2.26	12.91	2.27	2.58	14.68
860	Kharkov	Left on straw in field	Sept. 13	86.1	11.18	1.98	11.28	2.03	2.23	12.70
863	Weissenberg	Cut and left exposed	July 23	96.7	13.46	2.78	15.85	2.02	3.22	18.35
864	Weissenberg	Cut and protected	July 23	95.9	13.27	2.32	13.25	1.91	2.68	15.25
865	Weissenberg	Left on straw in field	Aug. 8	99.3	12.79	3.07	17.53	2.17	3.52	20.10
877	Currel	Cut and left exposed	July 22	87.9	13.25	2.06	11.73	1.84	2.38	13.52
878	Currel	Cut and protected	July 22	87.5	12.45	2.01	11.47	1.87	2.28	12.96
879	Currel	Left on straw in field	Sept. 13	87.6	13.11	2.07	11.78	1.90	2.38	13.55
874	Beloglina	Cut and left exposed	July 22	77.4	12.72	1.83	10.40	1.93	2.09	11.91
875	Beloglina	Cut and protected	July 22	94.8	12.97	2.11	12.06	1.44	2.44	13.86
876	Beloglina	Left on straw in field	Sept. 7	91.2	11.82	2.05	11.71	1.98	2.53	13.27
Average		Cut and left exposed	.....	69.1	.....	.....	.....	.....	.....	12.92
Average		Cut and protected	.....	75.3	.....	.....	.....	.....	.....	13.12
Average		Left on straw in field	.....	72.1	.....	.....	.....	.....	.....	13.52



The above results are not as consistent as could be desired in answering the main question involved in this experiment and no attempt will be made to interpret them as bearing upon this particular question. They do, however, further bear out the results discussed in the experiment on the effect of allowing the grain to stand on the straw in the field after reaching the hard dough stage, for it will be noted that in four out of six cases there was a larger percent of typical kernels in the late cut samples than in those early cut, and in one other case (no. 879) the percentage was essentially the same in the late cut sample as in that early cut. In the matter of total protein, *the late cut grain all carried a higher percentage than did those of the early cutting.*

The average results from this standpoint are shown below:

	Per cent typical kernels	Per cent total protein
Early cut .....	69.1	12.92
Late cut .....	72.1	13.52

#### THE EFFECT OF IRRIGATION UPON THE PROTEIN CONTENT OF WHEAT

The idea has been quite current among observant growers that whenever the rains extended late into the spring the quality of the grain of that season was materially reduced, that this had much to do with the wide seasonal differences in the quality of grain in California, and possibly was the main factor in causing such differences. This idea is quite in harmony with what has been observed in other experiments as to the effect of irrigation upon the quality of grain. No definite data being at hand as referring to conditions in California, in 1908-09 trials were made with six types of wheat at Davis to determine the effect of early and late application of water to growing wheat by planting these six types on uniform soil in rows at about the ordinary rate of seeding on three different plats. The plats received the following treatment so far as water was concerned:

Plat A received irrigation.

Plat B was irrigated in the rows once just after the grain was out of the boot.

Plat C was irrigated in the same manner twice, once at the same period in the plants' growth as in Plat B and again just after the grain set.

All plats, then, received the rainfall of the season and Plat B in addition had one irrigation and Plat C had two irrigations, the last one very late in its period of growth.

The wheats used in the experiment and their original compositions were as follows:

TABLE SHOWING THE ANALYSIS OF ORIGINAL WHEATS USED IN IRRIGATION TRIALS AT DAVIS, CAL.

No.	Per cent typical kernels	Number kernels in 10 grams	Per cent total protein	Per cent gliadin	Per cent ash
730/06	96.0	276	12.83	4.97	2.32
726/06	87.7	245	13.55	4.42	2.20
870/07	.....	227	15.92	4.27	2.02
1049/08	73.9	313	12.50	4.24	1.99
870/07	90.1	239	12.17	4.12	2.27
338a2	100.0	284	11.28	.....	2.13

The grains were seeded on December 7. They all came up with a good stand on December 28-31 and were harvested in two lots on June 24 and 30, there being but one or two days difference in the time of ripening between the irrigated and the non-irrigated plats.

The analysis of the several lots grown on each plat is shown in the following table:

## CUT EARLY

Lab. No.	Plat A—No irrigation					Plat B—One irrigation					Plat C—Two irrigations				
	Number					Number					Number				
	Per cent typical in 10 kernels	Per cent protein	Per cent gliadin	Per cent ash	Per cent ash	Per cent typical in 10 kernels	Per cent protein	Per cent gliadin	Per cent ash	Per cent ash	Per cent typical in 10 kernels	Per cent protein	Per cent gliadin	Per cent ash	Per cent ash
730	87.0	272	14.42	5.26	1.83	86.0	270	13.06	4.99	2.09	90.0	280	13.60	4.46	2.03
726	98.0	221	15.22	4.85	1.82	57.0	197	12.38	4.46	1.93	22.0	198	11.02	3.08	1.98
807a08	99.0	223	14.57	4.55	1.86	99.0	218	14.51	4.30	2.05	81.0	197	12.95	4.51	1.87
1049	-----	-----	-----	-----	-----	79.0	253	11.98	4.00	1.69	80.0	258	12.52	4.77	1.63
870	97.0	213	15.08	5.43	1.78	95.0	215	13.63	4.34	1.88	97.0	213	11.64	3.88	1.72
338a	95.0	260	13.52	5.41	2.00	90.0	258	13.12	4.94	2.10	95.0	262	14.88	4.00	1.78
Av.	95.2	239	14.56	5.10	1.86	82.3	235	13.11	4.51	1.96	77.5	234	12.77	4.12	1.84

## CUT LATE

Lab. No.	Plat A—No irrigation					Plat B—One irrigation					Plat C—Two irrigations				
	Number					Number					Number				
	Per cent typical in 10 kernels	Per cent protein	Per cent gliadin	Per cent ash	Per cent ash	Per cent typical in 10 kernels	Per cent protein	Per cent gliadin	Per cent ash	Per cent ash	Per cent typical in 10 kernels	Per cent protein	Per cent gliadin	Per cent ash	Per cent ash
730	81.0	265	15.76	5.93	1.99	90.0	283	15.48	4.88	1.98	80.0	285	13.63	4.89	2.08
726	88.0	193	13.38	4.28	1.86	93.0	184	14.37	5.07	1.93	91.0	185	13.97	4.34	1.83
807a08	99.0	220	14.08	5.54	1.96	99.0	203	14.57	5.29	1.83	93.0	215	14.26	4.76	1.78
1049	92.0	284	15.62	6.60	1.73	92.0	264	13.86	5.63	1.90	91.0	280	15.11	5.89	1.77
807/07	99.0	226	14.88	4.96	1.74	99.0	203	13.30	4.16	2.06	99.0	220	14.11	4.75	1.73
338a	99.0	260	15.25	5.77	1.82	94.0	275	15.08	5.60	1.80	79.0	270	13.18	4.81	1.91
Av.	93.0	241	14.83	5.35	1.85	94.5	235	14.44	5.12	1.92	88.8	242	14.04	4.91	1.85

Examining these results, it will be seen that in both the early and late cut lots Plat A, which received no late application of water, carried the highest average per cent of protein, and that Plat C, which had two water applications, carried the lowest. Averaging the early and late cut lots, the following figures hold:

	Per cent typical kernels	Number kernels in 10 grams	Per cent protein	Per cent gliadin	Per cent ash
Plat A	94.1	240	14.70	5.22	1.85
Plat B	88.1	235	13.78	4.81	1.94
Plat C	83.1	238	13.40	4.51	1.84

This shows a gradual decrease in both the protein and gliadin content as the moisture was increased, is in entire harmony with results cited elsewhere, and seems to show that either irrigation or late rains tend to lower the gluten content of wheat, and that this climatic factor is a very prominent, if not the most important one in causing seasonal variation in the grain.

Comparing the early cutting with the late cutting, it will be seen that while there is some fluctuation between corresponding samples in the two cuttings, the averages bear out the experiments cited in the earlier pages to the effect that no deterioration occurs from such late cutting, and as a matter of fact in this series of trials there was an actual increase in the protein content in the late compared with the early cutting.

#### THE EFFECT OF REDUCING THE ATMOSPHERIC TEMPERATURE, AT NIGHT UPON THE PROTEIN CONTENT OF WHEAT

In the season 1907-08 an attempt was made to reduce the temperature, at different stages of plant growth, on certain plats on which were seeded several types of grain and to compare the protein content on these plats.

The general<sup>a</sup> plan of this experiment consisted of seeding several types of wheat in rows upon adjacent small plats of uniform soil. The rows were seeded north and south, and to prevent the plats from receiving the early sun and to assist in holding down the temperature in the early morning across the south end of the plats cooled during the first period of growth, a board fence was erected sufficiently high to shade during the

morning hours about one half of each plat throughout the period of cooling. From the time the grains were well up until the spring rains ceased a layer of ice was spread on a loose frame and placed over one half of each plat, within four inches of the top of the plants, every night during the first half of the growing period of the plants. A portion of the north half of each plat was left uniced. To retain more effectively the cooled air during the night the iced portion of the plats was entirely covered by a piece of heavy canvas.

In this experiment two varieties of common wheats and five strains of durum wheats were used. The composition of each sample harvested is shown below:

TABLE 14.—SHOWING EFFECT OF REDUCING NIGHT TEMPERATURE UPON THE COMPOSITION OF WHEAT KERNELS

NOT COOLED						
Lab. No.	Name	Per cent typical kernels	Number kernels in 10 grams	Per cent protein	Per cent gliadin	Per cent ash
745B/08	Kubanka 1440	98.65	217	15.04	5.10	1.84
851/08	Kubanka 2221	99.8	201	15.19	5.09	1.92
869/08	Kubanka 2239	99.6	177	15.77	5.44	1.87
871/08	Velvet Don	99.5	205	15.44	5.31	1.96
872/08	Gharnovka	99.5	207	15.15	5.09	1.83
920/08	Red Fife	96.9	361	17.40	5.82	2.19
962/08	White Fife	100.0	323	16.49	5.65	1.97
Average		99.1	241	15.78	5.36	1.93

#### COOLED FIRST PERIOD

Lab. No.	Name	Per cent typical kernels	Number kernels in 10 grams	Per cent protein	Per cent gliadin	Per cent ash
745B/08	Kubanka	99.75	254	14.95	4.52	1.88
851/08	Kubanka 2221	99.70	233	15.31	4.96	1.23
869/08	Kubanka 2239	100.00	213	14.61	5.27	2.04
871/08	Velvet Don	99.94	229	15.87	5.32	1.86
872/08	Gharnovka	100.0	241	15.60	4.67	1.90
920/08	Red Fife	100.00	384	17.76	.....	2.26
962/08	White Fife	100.00	380	18.24	5.75	2.41
Average		99.9	276	16.05	5.08	1.98

## COOLED SECOND PERIOD

Lab. No.	Name	Per cent typical kernels	Number kernels in 10 grams	Per cent protein	Per cent gliadin	Per cent ash
745B/08	Kubanka 1440	100.0	217	15.24	5.39	1.96
851/08	Kubanka 2221	100.0	215	15.45	4.96	1.80
869/08	Kubanka 2239	99.8	203	15.80	5.43	1.90
871/08	Velvet D	99.9	246	15.53	4.25	1.90
872/08	Gharnovka	100.0	218	16.15	4.89	1.90
920/08	Red Fife	97.5	350	17.80	5.96	2.31
962/08	White Fife	99.4	339	17.69	6.29	2.13
Average		99.6	255	16.23	5.31	1.99

Collecting the averages for comparison, the figures are as follows:

Treatment	Per cent typical kernels	Number kernels in 10 grams	Per cent protein	Per cent gliadin	Per cent ash
Not cooled .....	99.1	241	15.78	5.36	1.93
Cooled First Period .....	99.9	276	16.05	5.08	1.98
Cooled Second Period .....	99.6	255	16.23	5.31	1.99

While the figures for individual analyses are slightly erratic, yet it appears that the general effect of reducing the temperature in each period tended to increase the total protein, and that the tendency was greatest when the night temperature was reduced in the second period of growth, for in the case of cooling during the first period of growth five out of the seven trials showed a higher percentage of total protein than those grown under normal conditions, and the average total protein was 16.05 per cent against 15.78 in the case of the uncooled lot.

In the case of the lot cooled during the second period of growth (after the grain was in the boot) all seven showed an increased protein content over the uncooled lot, the respective averages showing 17.23 against 15.79 per cent.

Further, the effect of the cooling seems to have been greater from reducing the temperature during the second period of growth than during the first, for five of the cases in this comparison show increased protein with an average protein content of 16.23 per cent against 16.05 per cent, and a gliadin content of 5.31 per cent against 5.08 per cent.

RELATION OF INCREASING THE SEVERAL AVAILABLE PLANT  
FOODS IN THE SOIL TO THE PROTEIN CONTENT OF  
WHEATS AT THE UNIVERSITY FARM,  
DAVIS, 1908-12, INCLUSIVE

The general effect upon the protein content of wheat from increasing the available nitrogen and other plant food elements in the soil at the University Farm at Davis has been made the subject of study for the past four years and the results are discussed below. In these experiments Little Club wheat has been used each year. In order that any cumulative effect which might accrue from the nitrogen and the other plant foods used might be apparent, the seed from each plat was seeded back upon the same plat each succeeding season.

The original seed used in these experiments was grown in 1906, and had the following composition:

	As analyzed	In dry matter
Moisture .....	11.28	.....
Total protein .....	12.12	13.66
Gliadin .....	4.38	4.93
Glutenin .....	6.38	7.19
Non-gluten proteids .....	1.36	1.58
Ash .....	1.62	1.82
Kernels in 10 grams ....	•226	
Bushel weight .....	59 lbs.	

For the four years 1908, 1909, 1910, and 1912 the average result was as stated in the table below. During the season of 1911 the land was under bare summer fallow on account of its foulness with wild oats, which accounts for the omission of that year.

During these trials the one-twentieth acre plats received the indicated quantities of fertilizer each season.

TABLE 15. SHOWING THE EFFECT OF VARIOUS FERTILIZER INGREDIENTS UPON THE PROTEIN CONTENT OF WHEAT KERNELS

Fertilizer applied	Per cent total protein	Per cent gliadin	Ash
1. Nitrate of soda, 5 lbs.; hydrate of lime, 132½ lbs. ....	*10.36	3.000	1.75
2. Hydrate of soda, 5 lbs. ....	*11.08	3.280	1.73
3. Hydrate of lime, 132½ lbs. ....	11.41	4.037	1.93
4. Check, no fertilizer ....	10.81	3.869	1.98
5. Nitrate of soda, 5 lbs.; sulphate of potash, 6 lbs. ....	10.99	3.816	1.95
6. Nitrate of soda, 10 lbs. ....	11.00	4.130	1.99
7. Check, no fertilizer ....	11.18	3.866	1.86
8. Nitrate of soda, 5 lbs.; superphosphate, 20 lbs. ....	11.02	3.855	1.82
9. Nitrate of soda, 10 lbs.; sulphate of potash, 6 lbs. ....	11.91	4.416	1.80
10. Nitrate of soda, 10 lbs.; superphosphate, 30 lbs. ....	10.93	3.728	1.83
11. Check, no fertilizer ....	9.99	3.793	1.87
12. Superphosphate, 20 lbs. ....	10.64	3.888	1.86
13. Sulphate of potash, 6 lbs. ....	10.82	4.054	1.81
14. Check ....	10.77	3.978	1.79
15. Nitrate of soda, 5 lbs.; sulphate of potash, 12 lbs. ....	10.94	3.806	1.82
16. Nitrate of soda, 5 lbs.; superphosphate, 30 lbs. ....	10.57	3.878	2.20
17. Nitrate of soda, 10 lbs.; superphosphate, 30 lbs.; sulphate of potash, 12 lbs. ....	10.76	3.870	1.90
18. Nitrate of soda, 5 lbs.; superphosphate, 30 lbs.; sulphate of potash, 6 lbs. ....	11.00	4.291	1.87
19. Nitrate of soda, 5 lbs.; superphosphate, 30 lbs.; sulphate of potash, 12 lbs. ....	11.35	4.703	1.90
20. Nitrate of soda, 5 lbs.; superphosphate, 50 lbs.; sulphate of potash, 6 lbs. ....	11.92	4.815	1.91
21. Check ....	10.58	4.312	1.82
22. Sulphate of potash, 6 lbs.; superphosphate, 30 lbs. ....	11.06	4.617	1.81
23. Dry blood, 7 lbs.; superphosphate, 30 lbs.; sulphate of potash, 6 lbs. ....	11.24	4.748	1.86
24. Legume, 1907-08; nothing, 1908-09; superphosphate, sulphate of potash, 1909-10 ....	12.27	5.720	1.87
25. Superphosphate, 30 lbs., sulphate of potash, 6 lbs., 1907-08; legume, 1908-09; nothing, 1909-10 ....	12.86	5.440	2.03

\* Plots 1 and 2 were discontinued in 1910 on account of building and plots 28 and 29 substituted.



TABLE 15—(Continued)

Fertilizer applied	Per cent total protein	Per cent gliadin	Ash
26. Sulphate of potash, 6 lbs., superphosphate, 30 lbs., 1908-09; nothing, 1907-08; legume, 1909-10 .....	12.51	5.287	1.71
27. Check, no fertilizer .....	11.23	4.692	1.93
28. Nitrate of soda, 5 lbs.; hydrate of lime, 132½ lbs. ....	*11.91	5.265	1.86
29. Nitrate of soda, 5 lbs. ....	*11.89	5.065	2.14

\* Plats 1 and 2 were discontinued in 1910 on account of building and plats 28 and 29 were substituted.

## THE EFFECT OF NITROGEN

Collecting the results as to the effect of nitrogen upon the protein content, the following figures hold:

Plats receiving nitrogen		Check plats	
Plat No.	Per cent protein	Plat No.	Per cent protein
1	10.36	3	11.41
2	11.08	4	10.81
Av. for 2 yrs.	10.67		11.11
Plat No.	Per cent protein	Plat No.	Per cent protein
9	11.91	5	10.99
6	11.00	7	11.18
5	10.99	13	10.82
10	10.93	12	10.64
17	10.76	19	11.35
10	10.93	12	10.64
23	11.24	22	11.06
Average	11.10		10.95

From the above it does not appear that increasing the available nitrogen content of the soil in these trials has had any material influence in increasing the nitrogen in the grain, nor has there been any cumulative effect shown by its use. This is shown by the individual cases as well as in the general averages. Considering individual cases, Plat 5 and Plat 6 may be compared, each receiving equal amounts of potash, while the former received also 100 pounds of nitrate of soda per acre, but it carried only .17 per cent more protein than the plat receiving no nitrate. Plat 6 received an application of 200 pounds of nitrate of soda,

while Plat 7 received no fertilizer, and yet the average protein content of the latter shows .18 per cent higher than the former. Further, in the case where a complete fertilizer was used, as in Plats 17 and 19, the former receiving double the quantity of nitrate of soda, the latter showed .59 per cent protein above the former. The results for the entire period show that increasing the available nitrogen of the soil had no general influence toward increasing the protein content of the wheats, either when used alone or when used in connection with the other plant food elements.

Comparing the effect of nitric nitrogen with organic nitrogen, as dried blood, in a complete fertilizer, the following results stand for the four years:

	1908	1909	1910	1912	Average
Plat 18—Nitric nitrogen .....	9.24	11.32	9.99	13.45	11.00
Plat 23—Organic nitrogen .....	10.28	10.77	10.85	14.08	11.49

This set seems to show a very slight increase due to the organic nitrogen, but it is so slight as not to affect in any material way the quality of the grain, and the variations are such as to render it more than probable that such variations as do occur are due to other causes, possibly moisture, rather than to the nitrate present.

#### THE EFFECT OF INCREASING THE NITROGEN CONTENT OF THE SOIL IN THE PRESENCE OF LIBERAL AMOUNTS OF PHOSPHORIC ACID

In these trials, nitrogen in the form of nitrate of soda was used in connection with an application of phosphates, as shown in the tabulation:

Nitrogen and phosphate plats		Check plats	
Plat No.	Per cent protein	Plat No.	Per cent protein
10	10.93	8	11.02
10	10.93	12	10.64
8	11.02	12	10.64
18	11.00	22	11.06
20	11.92	22	11.06
Average	11.16		10.88

The average difference shows .28 per cent, an amount which is too small to be a matter of consideration for practical improvement in the quality of the grain. Further, it will be noted that such differences as do occur did not run always in the same direction.

#### THE EFFECT OF PHOSPHATES

In these trials the superphosphate used was from treated rock and the amounts used are given in the scheme previously shown.

Superphosphate plats		Check plats	
Plat No.	Per cent protein	Plat No.	Per cent protein
12	10.64	11	9.99
8	11.02	2	11.08
10	10.93	6	11.00
16	10.57	2	11.08
20	11.92	18	11.00
19	11.35	15	10.94
22	11.06	13	10.82
Average	11.07		10.84

In general, the phosphates as here shown appear to have had a slight tendency toward increasing the total protein, but it is very slight on the average. In five cases out of seven, however, the phosphate plats carried somewhat higher protein than did the check plats. A more extended study, however, will be required before any definite conclusion can be reached.

#### THE EFFECT OF POTASH

In these trials the potash was universally used in the form of sulfate in the amounts indicated in the general tabulation.

Potash plats		Check plats	
Plat No.	Per cent protein	Plat No.	Per cent protein
5	10.99	2	11.08
9	11.91	6	11.00
13	10.82	14	10.77
15	10.94	5	10.94
19	11.35	18	11.00
Average	11.20		10.96

There is shown a slightly increased protein content on the potash plats over the corresponding checks. Three out of the five cases show a higher protein content on the potash plats than on the checks, and one shows the same percentage on both plats. The difference is so small in the average that it might easily be due to difference in the moisture content of the soil, but data along this line as relating to these plats are not at hand, and therefore it would be unwise to attempt to draw conclusions until such data are secured.

#### . THE EFFECT OF GREEN MANURE CROPS

Plats 23, 24, and 25 have had legumes grown upon them every third year and cereal crops the other year of the rotative period, with superphosphates and potash added the second year after the legume. Comparing the result from these plats for the short period the trials have been under way with the average of the check plats, and with the average of those on either side, the following results appear:

	Per cent protein
Plats with legumes .....	11.06
Average of all checks, no legume .....	11.02
Average of two nearest checks, no legume ....	10.90

It is not apparent that any material change in protein content has been effected by the treatment employed.

#### GENERAL CONCLUSIONS

The results presented in the foregoing pages seem to warrant the following conclusions:

First—There are important seasonal, varietal, and individual variations in wheat plants with respect to protein content.

Second—The principal factor causing the most pronounced variation in the protein content of wheats is climate, particularly the moisture supply in the later growing period of the crop.

Third—The tendency of wheat kernels to change from a glutenous to a starchy condition is not a constant one, but is mainly dependent upon the individuality of the plant and upon seasonal influences, particularly moisture supply in the latter part of the growing period of the plants.

Fourth—In wheats 100 per cent of which are entirely starchy there may be a reversion to an entirely glutenous condition in a single season, or the reverse may occur, dependent upon the seasonal condition.

Fifth—Allowing the grain to stand on the straw in the field until fully ripe does not materially affect the protein content.

Sixth—The protein content of wheat is affected by the time of seeding, the product of late seeded grain having a higher percent of protein than that of early seeded grain.

Seventh—The protein content of wheat is very largely influenced by the water content of the soil in the later period of its growth, and the effect of either irrigation or rainfall during this period is to lower its protein content.

Eighth—The percentage of sunshine which the grain receives during its period of growth has a somewhat direct bearing upon its protein content, but other seasonal conditions are more important.

Ninth—Retarding the growth through cooling the atmosphere has a tendency to increase the protein content.

Tenth—The quantity of available nitrogen in the soil either alone or in the presence of other available plant foods, provided there be sufficient to supply normal growth, appear to have little, if any, influence upon the protein content.

Eleventh—The low gluten content of wheats grown in California is not due to soil exhaustion, but rather to the following causes: (1) To climatic factors which allow a long growing period; (2) to relatively early seeding; (3) to the use of varieties inherently low in gluten; (4) to a lack of selecting highly glutenous seed.

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**THE EFFECT OF COPPER, ZINC, IRON AND  
LEAD SALTS ON AMMONIFICATION  
AND NITRIFICATION IN SOILS**

BY

**C. B. LIPMAN AND P. S. BURGESS**

**UNIVERSITY OF CALIFORNIA PRESS**  
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THE EFFECT OF COPPER, ZINC, IRON AND  
LEAD SALTS ON AMMONIFICATION  
AND NITRIFICATION IN SOILS

BY

C. B. LIPMAN AND P. S. BURGESS

In the course of investigations on the effect of smelter wastes on crop growth one of the writers decided to test also the effects of the salts of the heavy metals on the transformation of organic nitrogen in the soil into ammonia and nitrates which serve as the source of nitrogen for plants. This correlation between the effects of external factors upon both the soil flora and the physiological condition of plants was deemed eminently worth while, for reasons which are at once obvious to the careful student of soil fertility problems. Some of the results obtained with plants have been published,<sup>1</sup> and still fuller data dealing with the same problem will soon find publication elsewhere. In view of the foregoing the writers have carried out experiments dealing with the toxic and stimulating effects of copper, zinc, iron and lead on the ammonifying and nitrifying flora of a sandy soil. The results, owing to their interesting nature and their eogeneity at this time are here given apart from the results obtained with plants.

METHODS OF THE EXPERIMENTS

The work here described was carried out by the direct soil-culture method in a manner fully explained by one of us elsewhere.<sup>2</sup> No attempt was made to modify the soil flora, but the

<sup>1</sup> Bot. Gaz., vol. 55, p. 409.

<sup>2</sup> Cent. für Bakt., 2<sup>te</sup> Abt., vol. 32, p. 58, vol. 33, p. 305.



soil with its natural flora was employed. The ammonia determinations were made in accordance with methods described in the papers above cited and the nitrate determinations in accordance with the method described by Burgess.<sup>3</sup>

The work carried out by other investigators dealing with the subject in hand has thus far been rather meager and but few of the results possess pertinence with respect to our findings. In studying the amounts of copper in vineyard soils due to accumulation from copper-containing sprays, Prandi<sup>4</sup> was unable to note any damage to vines after the copper sprays had been used for a number of years. Moreover, he did not adjudge dangerous the amounts of copper which had accumulated in the soils studied in the quantities in which he found it there. He does, however, make the further interesting speculative statement that copper may have an important influence on the soil organisms. The original work of Eickmeyer<sup>5</sup> is unfortunately not accessible to us and, while it may contain some of the most cogent information on our subject which is in print, we can ascertain only that the investigator named studied the effects of copper and iron sulfates, among other poisons, on soil bacteria. In studying the bacteriology of ammonium compounds Ehrenberg<sup>6</sup> observes that the cause of the difference in the effects of ammonium compounds used for fertilizers in zinc pots and in the field must be that the zinc exercises a deleterious effect on "ammonia-fixing" bacteria which compete with the plants for soluble nitrogen in the soil. Owing to that, he believes ammonium fertilizers stimulate plants in zinc pots but have no effect in the field. Among studies on the effects of the poisonous metals here considered, on some of the lower organisms, may be mentioned those of Latham,<sup>7</sup> who obtained stimulation with low concentrations of  $\text{ZnSO}_4$  for *Aspergillus niger*, and those of Remy and Rosing,<sup>8</sup> which indicate a marked stimulating effect of iron on *Azotobacter* organisms.

<sup>3</sup> Univ. Calif. Publ. Agr. Sci., vol. 1, no. 4, p. 51.

<sup>4</sup> Staz. Sper. Gr. Ital., vol. 40, p. 531, cited from E. S. R., vol. 19, p. 755.

<sup>5</sup> Winner Landw. Ztg., vol. 57, p. 600, cited from E. S. R., vol. 19, p. 625.

<sup>6</sup> Fuhling's Landw. Ztg., vol. 27, p. 449.

<sup>7</sup> Cited from E. S. R., vol. 21, p. 421.

<sup>8</sup> Cent. für. Bakt., 2<sup>te</sup> Abt., vol. 30, p. 349.

The only work with which the writers are acquainted which allows of any direct comparison with our results is included in the splendid series of investigations of Fred<sup>9</sup> on the effects of small or limited quantities of various poisons on the higher as well as the lower plant organisms. In addition to showing the stimulating powers of certain poisons like  $\text{CS}_2$  and  $\text{C}_2\text{H}_5 \rangle \text{O}$  on the bacterial count, nitrogen-fixing power, etc., of soils in direct soil cultures, that investigator gives evidence also of work done with the higher plants yielding similar results. Of a series of metallic salts, further, with which Fred worked he was unable to demonstrate stimulation to nitrification except perhaps one case of slight stimulation through the use of  $\text{FeSO}_4$  (100 mgs. per 100 grams of soil). The salts tested were  $\text{MnSO}_4$ ,  $\text{FeSO}_4$ ,  $\text{CuSO}_4$ , and  $\text{NaCl}$ . The nitrifiable material employed was  $(\text{NH}_4)_2\text{SO}_4$  and not the organic nitrogen of blood which we used.

Aside from these investigations, only one or two of which have any direct bearing on our subject, but little has been accomplished of either direct or indirect applicability to our results. Where any studies were carried out dealing with the effects of the heavy metals on bacteria, such as those of Kellerman and Beckwith<sup>10</sup> or Jackson,<sup>11</sup> they were prosecuted in solutions and therefore, as one of us has emphatically shown in other publications,<sup>12</sup> are scarcely comparable with results from soil cultures such as are below discussed, or with results obtained with plants in soil cultures.

Since the experiments below described were completed there has appeared from the pen of Greaves<sup>13</sup> a paper bearing on the effects of arsenic on ammonification and nitrification in soils. This work, because of the methods employed, is of pertinence here, and interesting, because of the stimulating powers of considerable quantities of arsenic in soils to ammonia and nitrate production by the natural flora. While arsenic, chemically considered, is a

<sup>9</sup> Cent. für Bakt., 2<sup>te</sup> Abt., vol. 31, p. 185.

<sup>10</sup> Bull. 100, Bur. Pl. Ind., U. S. D. A.

<sup>11</sup> Jour. Am. Chem. Soc., vol. 27, II, p. 675.

<sup>12</sup> Bot. Gaz., vol. 55, p. 409 and literature there cited.

<sup>13</sup> Cent. für Bakt., 2<sup>te</sup> Abt., vol. 39, p. 542.

totally different substance from the metals we have studied, it is interesting to compare its effects on the soil flora with those exercised by copper, zinc, lead and iron. This is especially so since arsenic so frequently occurs with the other elements in soils and in smelter wastes from which it may be transported to agricultural soils.

In closing this brief review of investigations dealing with the subject of this paper or one allied thereto, it should also be added that some fragmentary information was obtained in the course of other investigations by J. G. Lipman and his associates<sup>14</sup> which refers to the effects of  $\text{CuSO}_4$  and  $\text{FeSO}_4$  among other compounds on the ammonifying flora of the soil. These results indicate that copper stimulates ammonification very slightly at a concentration of 0.1 per cent, that  $\text{ZnSO}_4$  gives no stimulation and is toxic at all concentrations, and that  $\text{FeSO}_4$  at the concentration of 1 mg. per 100 grams of soil gives marked stimulation.

#### THE AMMONIFICATION EXPERIMENTS

To a fifty-gram portion of dry soil, one gram of tankage (9.62% N) was added and thoroughly mixed with it. Sterile distilled water was then added, to make optimum moisture conditions, and also varying quantities of the salts to be tested. The soil, water and salt mixture was thoroughly stirred in the tumbler, the latter covered with a Petri dish cover and incubated for one week at 27° C to 30° C. The results of the ammonia determinations at the end of the incubation period, together with other necessary explanatory data, will be found in Table 1. The sulfates of the metals tested were employed throughout. It will be noted that the results of duplicate determinations are given throughout, averages being omitted since the reader can so readily ascertain them from the data submitted.

It is at once obvious, from even a casual examination of the data given in Table 1, that none of the metals employed exercises a stimulating effect on the ammonifying flora of the soil employed even at the smallest concentrations (50 p.p.m.). Despite that

<sup>14</sup> N. J. Exp. Sta. Bull. no. 246, p. 32.

TABLE 1

THE EFFECT OF CU, ZN, FE, AND PB ON AMMONIFICATION IN SOILS

No.	CuSO <sub>4</sub> % of dry soil	N as NH <sub>3</sub> produced mgs.	ZnSO <sub>4</sub> % of dry soil	N as NH <sub>3</sub> produced mgs.	FeSO <sub>4</sub> % of dry soil	N as NH <sub>3</sub> produced mgs.	PbSO <sub>4</sub> % of dry soil	N as NH <sub>3</sub> produced mgs.
1	0	40.60	0	40.60	0	40.60	0	40.60
2	0	39.29	0	39.20	0	39.20	0	39.20
3	.005	40.32	.005	36.82	.005	39.34	.005	38.50
4	.005	38.50	.005	36.68	.005	37.94	.005	37.94
5	.010	36.40	.010	39.20	.010	39.48	.010	37.52
6	.010	lost	.010	37.38	.010	37.38	.010	38.78
7	.025	35.00	.025	36.82	.025	37.24	.025	41.58
8	.025	35.70	.025	35.98	.025	lost	.025	39.90
9	.050	35.98	.050	33.74	.050	37.10	.050	40.32
10	.050	36.26	.050	32.20	.050	39.20	.050	38.78
11	.075	32.20	.075	31.50	.075	36.68	.075	39.76
12	.075	30.80	.075	31.92	.075	37.94	.075	38.92
13	.100	29.96	.100	31.08	.100	38.22	.100	35.70
14	.100	28.00	.100	29.12	.100	<del>38.34</del>	.100	35.84
15	.125	27.58	.125	31.50	.125	35.98	.125	35.00
16	.125	28.84	.125	31.50	.125	34.58	.125	35.42
17	.150	28.70	.150	34.30	.150	35.56	.150	33.04
18	.150	29.96	.150	33.74	.150	37.52	.150	34.72
19	.200	26.66	.200	34.58	.200	36.40	.200	35.00
20	.200	27.44	.200	33.60	.200	37.10	.200	32.90
21	.250	28.70	.250	32.90	.250	36.68	.250	35.00
22	.250	27.58	.250	32.06	.250	37.52	.250	36.26

fact, however, the toxicity of no one of them is very marked. While this latter observation is in accord with the findings of J. G. Lipman above referred to, the former result is entirely at variance with them. It is significant to remark in this connection, however, that the agreement between duplicate determinations was on the whole much better in our experiments than in those carried out at the New Jersey Experiment Station and should therefore render our results more trustworthy. Indeed, as the authors of the New Jersey bulletin state, for their work "there is not sufficient evidence upon which to base conclusions."

Comparing the relative toxicities of the salts tested by us it would appear that copper is the most toxic, zinc the next, lead the next, and iron the least toxic of the four sulfates. This assertion is based on the fact that at the highest concentrations at which it was used, copper depresses ammonification by about

30 per cent, zinc by 20 per cent, lead by about 15 per cent, and iron by only about 12 per cent of that of the normal yield. Despite this fact, however, copper begins to manifest its toxicity most slowly of the four metals, since it shows little if any toxicity at a concentration of .005 per cent, whereas all the other metals show clearly a slight toxic effect even at that concentration. Just why this peculiar relationship between the metals tested and the ammonifying flora should obtain is by no means easy of explanation.

Speaking of the data in Table 1, by and large, it seems justifiable to venture the assertion that all the metals become markedly more toxic at concentrations of 0.1 per cent of the dry weight of the soil or in concentrations in excess of that amount. Other details worthy of mention here are the fact that at times the depressing effect of a certain concentration is followed by a stimulating effect at an increased concentration of the same metallic salt. Also, the very striking fact is noted of the relatively slight increases of toxicity which accompany relatively large increases in concentration of the salt. The latter is particularly marked in the case of  $\text{FeSO}_4$ . Whether or not this latter result is to be explained by the differences in solubility which obtain between the different metallic sulfates employed, as well as by the different adsorptive power exercised by the soil towards every one of them, still remains to be shown.

In general, it is worthy of special remark here that though these so-called very toxic metals manifest undoubted toxicity towards the ammonifying flora, and no stimulation in any concentration of them, the toxic effect is relatively small and in some cases even absolutely slight.

#### THE NITRIFICATION EXPERIMENTS

To 100-gram portions of sandy soil in tumblers were added two grams of dried blood (13.29% N), enough water to make optimum moisture conditions, and the necessary amounts of the salts to be tested. The mixture was prepared for incubation as above described for the ammonification cultures and incubated

for four weeks at 27° C to 30° C. Other explanatory data are given in Table 2, which shows the results obtained from the nitrate determinations made as above explained.

TABLE 2  
THE EFFECT OF CU, ZN, FE, AND PB ON NITRIFICATION IN SOILS

No.	CuSO <sub>4</sub> % of dry soil	Nitrates produced mgs.	ZnSO <sub>4</sub> % of dry soil	Nitrates produced mgs.	FeSO <sub>4</sub> % of dry soil	Nitrates produced mgs.	PbSO <sub>4</sub> % of dry soil	Nitrates produced mgs.
1	0	10.64	0	11.20	0	11.20	0	11.20
2	0	9.80	0	10.64	0	10.64	0	10.64
3	.0125	7.84	.0125	10.36	.0125	14.56	.0125	5.60
4	.0125	10.64	.0125	11.48	.0125	16.24	.0125	7.00
5	.0250	11.20	.0250	11.20	.0250	12.60	.0250	9.52
6	.0250	10.08	.0250	12.60	.0250	12.88	.0250	8.68
7	.0500	17.64	.0500	16.24	.0500	13.72	.0500	10.08
8	.0500	18.76	.0500	17.36	.0500	13.44	.0500	11.20
9	.0750	24.36	.0750	21.00	.0750	14.00	.0750	11.48
10	.0750	24.92	.0750	23.80	.0750	11.76	.0750	14.00
11	.1000	23.80	.1000	23.52	.1000	8.40	.1000	7.56
12	.1000	24.64	.1000	22.68	.1000	7.84	.1000	7.56
13	.1250	21.00	.1250	17.08	.1250	18.20	.1250	11.20
14	.1250	19.32	.1250	17.22	.1250	21.00	.1250	12.88
15	.1500	20.44	.1500	17.08	.1500	20.72	.1500	6.44
16	.1500	19.88	.1500	17.36	.1500	19.60	.1500	6.72

It is quite clear from the foregoing table that all of the salts used exercise marked effects on the nitrifying flora of the soil employed. For the reason that some pronounced differences obtain between the action of the different salts we shall discuss each of them separately at first.

#### CuSO<sub>4</sub>

It can be seen at a glance that the data in Table 2 show copper to be the most stimulating of the salts employed so far as a soil-nitrifying flora is concerned. While, unfortunately, the duplicate determinations for the lowest concentration do not agree well, they indicate clearly that in those quantities (0.0125 per cent. or 125 parts per million) copper has either no effect on the nitrifying flora or a slightly depressing one. By doubling the concentration of the copper salt we obtain a slightly stimulating effect, but by doubling the latter concentration we obtain

a most striking stimulating effect on the nitrifying flora, which results in the production of nearly twice as much nitrate nitrogen as is yielded in the normal soil receiving no copper at all. But the stimulation of the nitrifying flora does not cease at a concentration of  $\text{CuSO}_4$  equivalent to 0.05 per cent of the dry weight of the soil. Even at a concentration of 0.075 per cent of  $\text{CuSO}_4$  we obtain stimulation and one-third again as much nitrate as is produced when copper is present at the preceding concentration (0.05 per cent  $\text{CuSO}_4$ ). At 0.1 per cent  $\text{CuSO}_4$  about the same amount of nitrate is produced as at a concentration of 0.075 per cent, or perhaps slightly less, and at the highest concentration of  $\text{CuSO}_4$  employed, namely 0.15 per cent, we obtain an increase of about 100 per cent of nitrate over the normal soil to which no copper is added. That such remarkable stimulation should be exercised by  $\text{CuSO}_4$  for the nitrifying flora and at such high concentrations of that supposedly toxic salt is a new and interesting fact of great significance.

While our results agree with those of Fred above cited as regards the lower concentration of  $\text{CuSO}_4$  which he employed, namely 0.01 per cent, they are wholly at variance with his results as regards his only other concentration, 0.1 per cent  $\text{CuSO}_4$ . The latter gave Fred no stimulation for the nitrifying flora whatever, but it yields us nearly the maximum point of stimulation of  $\text{CuSO}_4$  for the flora in question, and in a series of results in which the existence of stimulation is throughout remarkable. To what circumstance these differences between our work and that of Fred are attributable is not readily determined. It can scarcely be due to the difference in the strain or vigor of the nitrifying flora, for we have obtained similar results to those above described with widely different soils which were employed in corresponding vegetation experiments. That the nitrifiable materials were wholly different may, however, be a factor of considerable significance. Not only because of its totally different physical nature, but because of possible antagonistic effects which it might induce,  $(\text{NH}_4)_2\text{SO}_4$  might well be expected to yield different results in nitrification work from those obtained with dried blood or similar forms of nitrogen, especially when salt effects are studied.

A question more difficult of solution is that involving the cause or causes of the stimulating effects of  $\text{CuSO}_4$  on the nitrifying flora or the production of nitrates in soils. The experiments of Fred have included such as aimed at the discovery of the reply to the question just raised. The results of such experiments, however, would seem to throw no positive light on its solution. Two factors do seem, nevertheless, to have been eliminated. But little support was found for the idea that poisons like the salts above employed destroy soil toxins which might interfere with bacterial efficiency, as well as for the further idea that the effect of such poisons in the destruction of amoebae might manifest itself by a stimulation for the soil organisms. It seems, to use Fred's expression, to be a "Reizwirkung" on the part of the salts and other poisons added to the nitrifying flora, but the question as to how this "Reizwirkung" is accomplished still remains unanswered. This question is further discussed below.

#### $\text{ZnSO}_4$

Zinc evidently exercises relatively the same influence on the nitrifying flora as copper. Like the latter, it stimulates but very slightly at a concentration of 0.0125 per cent  $\text{ZnSO}_4$ , does so very definitely at a concentration of 0.025 per cent, very markedly at 0.05 per cent, and still more markedly at concentrations of 0.075 per cent and 0.10 per cent, at which latter the highest point of stimulation is reached. Concentrations in excess of 0.1 per cent  $\text{ZnSO}_4$  seems to be much less stimulating to nitrate production than similar amounts of  $\text{CuSO}_4$ . The absolute amounts of nitrate produced by the nitrifying flora under the stimulus of zinc salts seem to be throughout slightly smaller than those produced by similar flora in the presence of  $\text{CuSO}_4$ .

#### $\text{FeSO}_4$

With  $\text{FeSO}_4$  we obtain effects on the nitrifying flora which possess many characteristics different from those obtained with  $\text{CuSO}_4$  and  $\text{ZnSO}_4$ . In the first place, stimulation is marked with the smallest concentration, 0.0125 per cent  $\text{FeSO}_4$ , while it is either extremely slight or wanting with the other salts at



similar concentrations. While stimulation continues up to and including concentrations of 0.075 per cent, it is not so great as with the smallest concentration, and increases or decreases with the different amounts of  $\text{FeSO}_4$  employed, with no regularity. Then, beyond the concentration of 0.075 per cent  $\text{FeSO}_4$ , a wholly inexplicable sudden toxicity manifests itself, and stranger still, beyond that point with two additional and larger concentrations employed we obtain the most marked stimulation in the whole series of cultures and about equivalent to the stimulation effected by  $\text{CuSO}_4$  at similar concentrations.

The causes of this erratic behavior of  $\text{FeSO}_4$  are a mystery as yet. We suggest that the rapidity of oxidation of the  $\text{FeSO}_4$  may vary and so the ferrous and ferric salts may yield different effects, though this explanation does not appear wholly satisfactory. The distribution of the salt in the soil culture may also be a factor. What appears to us as another promising explanation for the behavior of the  $\text{FeSO}_4$  under discussion is its effect on the physical condition of the soil and the modification of the latter's absorptive power for moisture and gases. Whether or not the effects noted may be related to transformations in the amounts of available bacterial foods present cannot be answered from the data in hand.

### $\text{PbSO}_4$

In this series of cultures we are confronted with effects which are in certain definite ways different from either those of  $\text{CuSO}_4$  and  $\text{ZnSO}_4$ , on the one hand, and  $\text{FeSO}_4$  on the other hand. Lead is the only one of the metals used above which exercises an unquestionable and marked toxicity on the nitrifying flora at even the lowest concentration (0.0125 per cent  $\text{PbSO}_4$ ). But while that is so, larger amounts of  $\text{PbSO}_4$  make conditions in the soil more favorable for nitrification as the lead salt increases in concentration from 0.0125 per cent to 0.05 per cent, at which point nitrification is about as active as in the normal soil. When, however, the  $\text{PbSO}_4$  is still further increased we obtain what appears to be a definite stimulation to nitrification. Oddly enough, however, at a concentration of 0.1 per cent,  $\text{PbSO}_4$

behaves like a similar concentration of  $\text{FeSO}_4$  to the nitrifying flora and becomes about as toxic as 0.0125 per cent was shown to be above. Then at 0.0125 per cent  $\text{PbSO}_4$  the latter again behaves like  $\text{FeSO}_4$  in that definite stimulation follows the toxicity of the previous concentration, but while the stimulation of the  $\text{FeSO}_4$  at a similar concentration is very marked it is only slight in the case of  $\text{PbSO}_4$ . Then, again, at the further concentration of 0.15 per cent  $\text{PbSO}_4$ , the latter acts entirely differently from all the other salts in that it becomes markedly toxic again.

The idiosyncracies of  $\text{PbSO}_4$  as regards the nitrifying flora of a soil are even more numerous than those of  $\text{FeSO}_4$ . However, it must be borne in mind that  $\text{PbSO}_4$  is a very insoluble salt and while the other salts are capable of being added in solution to the cultures,  $\text{PbSO}_4$  has to be mixed with the soil as thoroughly as possible in its dry state. This fact would account for a poor distribution of the salt through the soil mass and thus indicate one possible cause of the extremely erratic behavior of  $\text{PbSO}_4$  in nitrification cultures. The fact, however, that  $\text{FeSO}_4$  and  $\text{PbSO}_4$  behave alike at a concentration of 0.1 per cent would seem to be more than a mere coincidence, and some of the speculations made above with respect to  $\text{FeSO}_4$  and its behavior may have some cogency here. The most striking fact, however, gleaned from the series of cultures with  $\text{PbSO}_4$  is that the latter possesses a stimulating power toward the nitrifying flora. Considering the well-known toxic powers of lead with respect to other organisms, this fact assumes considerable importance. In all cases, of course, the stimulating powers of the other salts are much greater than that of  $\text{PbSO}_4$ .

#### GENERAL DISCUSSION

The experimental data above submitted constitute the first series of systematic experiments dealing with the effects of copper, zinc, iron and lead on the ammonifying and nitrifying flora of the same soil. Once more we are confronted by the striking differences between the physiological characteristics of the ammonifying and the nitrifying flora of soils. Other similar

cases, though not so marked, have been commented on elsewhere by one of us (see literature above cited). While the sulfates of all the metals named are toxic to ammonification, and in very low as well as very high concentrations, ranging from 50 parts per million to 2500 parts per million, they manifest a relatively low toxicity for the flora concerned in that phase of nitrogen transformation. On the other hand, all the same metals exercise, in a large variety of concentrations, a marked stimulating effect on the nitrifying flora. That differences of such magnitude should exist between two flora of the soil, one being presumably dependent for its raw material on the other, is, to say the least, amazing and not wholly explicable. Since, however, nitrification in the broader sense of the term represents the algebraic sum of the activities of all forms of nitrogen-transforming bacteria in the soil, it would appear from our results that the net effect of the activity of the metallic salts tested is to insure a larger nitrate supply in the soil. This must be so, since in the most extreme cases above studied ammonification is depressed only to an extent which lowers the total ammonia yields below that of the salt-free soil by about 30 per cent, whereas nitrate production is frequently enhanced in efficiency by the presence of the same metals to an extent which doubles the yield of a normal soil flora.

Our findings would therefore render much easier the explanation of the oft-noted stimulating effect of copper, in particular, and metallic salts in general (in proper concentrations), on the higher plants, regarding which, also, we have accumulated considerable data. If a larger nitrate production in the soil follows the application of a metallic salt as above shown, the nitrogen nutrition of plants must go on with greater facility and adequacy; hence increased growth.

As above intimated, the underlying causes of our very interesting results are not easy to discover. In addition, however, to the speculations on that subject which we make, and which we recognize for more reasons than one as far from satisfying, we have obtained some experimental evidence on the point in question which possesses more cogency. It appears from these that water absorption is hastened by germinating seeds and young

plants in the soil in the presence of copper. Why might not a similar effect be exercised by metals as regards the nitrifying bacteria which, physiologically speaking, in every way resemble the higher plants much more than the other soil flora?

### CONCLUSIONS

1. Copper, zinc, iron and lead exercise toxic effects on the ammonifying flora of a sandy soil from Southern California in all concentrations, ranging from 50 to 2500 parts per million of their sulfates.

2. Such toxicity is relatively small, however, and is more marked at concentrations below 0.1 per cent than above it, in most cases.

3. The metals named exercise no stimulating effect on the ammonifying flora at any concentration.

4. Copper, zinc, iron and lead exercise marked stimulating effects on the nitrifying flora of the same soil and frequently more than double the normal nitrate yield.

5. The same metals may in very small concentrations exercise toxic effects on the nitrifying flora or no effect at all, but they are markedly stimulating at much higher concentrations.

6. With the one exception of lead sulfate, the metals named showed very marked stimulation for nitrification even at 0.15 per cent, the highest concentration employed.

7. Comparisons are given of our work with that of one or two other investigators on portions of the same subject. In some cases our results confirm previous work. In other cases they are wholly at variance with it.

8. A brief discussion is given of possible causes for the effects of the metallic salts under consideration on the soil flora.

9. Fuller consideration will be given in another paper to the theoretical phases of the subject treated above.

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**STUDIES ON AMMONIFICATION IN SOILS  
BY PURE CULTURES**

**BY**

**C. B. LIPMAN AND P. S. BURGESS**

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**STUDIES ON AMMONIFICATION IN SOILS**  
**BY PURE CULTURES**

BY

C. B. LIPMAN AND P. S. BURGESS

The study of the physiological efficiency of soil bacteria rather than their number is admittedly the dominant method in soil bacteriological investigations. In view of this fact it is singularly striking to note how little work has been accomplished in the study of some phases of the physiological efficiency of pure cultures of certain groups of soil bacteria. This is especially so, since the introduction of radical changes in our soil bacteriological methods have made it necessary to repeat some, if not all, of the work which had been carried out by the old methods. However this may be, it remains a fact that, since the publication of Marchal's' splendid work on ammonification in solution cultures, but scant information has been adduced from studies of soil bacteria which relate to the physiology as well as the physiological efficiency of even the more common ammonifying bacteria. Certainly, the work carried out along this line in direct soil cultures, which recent work has shown to be so far superior to solution cultures, has been very meager indeed.

In order, therefore, to glean some useful information relative to the physiological efficiency of pure cultures of a number of ammonia-producing bacteria, the writers deemed it wise to select a number of organisms and to compare their power to produce ammonia not only from one form of organic matter, but from several forms, most of which have found use in farm practice as fertilizers. Accordingly, the following organisms in pure

culture were selected for the experiments: *B. mesentericus vulgaris*, *Ps. putida*, *B. vulgaris*, *B. megatherium*, *B. mycoides*, *B. subtilis*, *B. tumescens*, *Sarcina lutea*, *B. proteus vulgaris*, *B. icteroides*, *B. ramosus*, *Streptothrix*, sp., *Ps. fluorescens*, *B. vulgaris* (Novy strain), *Mic. tetragenus*. The organic materials experimented with were dried blood (13.16% N), tankage (9.62% N), cottonseed meal (5.5% N), sheep and goat manure (2.13% N), peptone (14.14% N), fish guano (8.63% N), and bat guano (3.96% N).

#### METHOD OF EXPERIMENTS

Fifty-gram portions of soil were placed in tumblers and thoroughly mixed with the organic material to be tested. The tumblers were covered with Petri dish covers and sterilized in the autoclave at a pressure of thirty pounds for three hours. After cooling, the soils were each inoculated with a 1 cc. suspension of the organisms to be tested, made up by shaking with some sterile water a young slope culture grown on bouillon agar. The soil was then stirred with a sterile spatula after enough sterile water had been added to make a moisture content in the soil about equal to the optimum. The soil cultures thus prepared were incubated at 28° to 30° C for twelve days. After the incubation period the soils were transferred to copper distilling flasks, 400 cc. of distilled water and an excess of Mg O added, and distilled into standard H/10 HCl. The ammonia was then determined in the usual way.

No attempt was made to run all the series with the different forms or organic matter at the same time, because only the relative powers of the different organisms to produce ammonia were sought. For the same reason amounts of organic matter were chosen in the different series which would least affect the physical conditions obtaining in the cultures rather than amounts employed which would make the total amount of nitrogen added the same in all series.

For the reasons above given, therefore, the effects of the various organisms on any given form of organic material will be treated below as a separate series in the case of each soil and



comparisons between the different series made only where permissible. The data, moreover, are presented so that the duplicate determinations which were carried out in all cases may be compared. The averages of duplicate determinations, however, are also given as well as the percentages of nitrogen in the organic matter which was transformed to ammonia.

Three different California soils were tested with each one of the ammonifiable materials. The soils were a sandy soil from Anaheim, a clay loam from Davis, and a black clay-adobe soil from Berkeley. The mechanical and chemical analyses of these soils are given in Tables I and II which follow.

TABLE I  
MECHANICAL ANALYSES OF SOILS

Hyd. value	Sandy	Clay-Loam	Clay-Adobe
Clay	5.78	19.12	31.93
0.25 mm.	14.59	40.93	25.77
0.50	1.04	3.35	3.40
1	2.30	6.60	4.77
2	5.28	7.75	7.49
4	9.62	8.78	6.20
8	11.58	8.10	.87
16	4.87	3.30	2.78
32	15.23	4.15	7.66
64	29.40	3.07	5.44

TABLE II  
CHEMICAL ANALYSIS OF SOILS

	Sandy	Clay-Loam	Clay-Adobe
Insoluble matter	73.59	53.55	} 77.84
Soluble silica	11.17	19.77	
K <sub>2</sub> O	.64	.75	.45
Na <sub>2</sub> O	.15	.11	.07
CaO	1.39	.82	1.05
MgO	.93	1.39	1.21
Mn <sub>2</sub> O <sub>4</sub>	.04	.04	.08
Fe <sub>2</sub> O <sub>3</sub>	5.10	7.56	4.68
Al <sub>2</sub> O <sub>3</sub>	3.92	10.04	7.79
P <sub>2</sub> O <sub>5</sub>	.12	.13	.23
SO <sub>3</sub>	.02	.03	.08
Water and organic matter	2.88	5.62	5.72

The pure cultures of the organisms employed in these investigations were obtained by one of us from the bacteriological laboratories of the University of Illinois. Fresh strains of *B. subtilis* and *B. mycoides* were, however, isolated by us from California soils in order to check the stock cultures of the same organisms. The same relative powers of producing ammonia were, however, found to obtain with the fresh as with the old, and different strains thus strengthening the validity of the results below discussed.

#### SERIES I. EXPERIMENTS WITH DRIED BLOOD

##### *Sandy Soil*

Two per cent of finely sifted dried blood was added to the soils in this series, or one gram per fifty grams of soil. In the case of the sandy soil three series in duplicate were tried, and the same relation under different conditions having been found to obtain between the ammonifying powers of the different organisms, we give only one duplicate set of the determinations. The incubation period was twelve days throughout at a temperature varying between 27° C and 30° C. The results of the ammonification determinations with all the soils using dried blood throughout are given in Table III. The percentage of nitrogen of the total amount added which is made available is also given in every case.

The data in Table III most strikingly indicate the superiority of *Ps. putida*, *B. vulgaris* and *Sarcina lutea* to all other organisms in their efficiency at the production of ammonia from the nitrogen of dried blood. The next fact of singular interest is that *B. mycoides*, which has, in the hands of several investigators, shown such marked superiority over other ammonifying organisms in the production of ammonia from organic nitrogen in solutions, manifests a relatively low power to transform the nitrogen of dried blood in soil cultures into ammonia. This, moreover, cannot be considered accidental, since different strains of *B. mycoides*, as above explained, exhibited that same lack of vigor in three duplicate sets of cultures run at different times and under slightly varying conditions of temperature and period of incuba-

TABLE III  
AMMONIFICATION OF DRIED BLOOD

Name of organism	SANDY SOIL				CLAY-LOAM SOIL				CLAY-ADORE SOIL			
	Mgs. N. as NH <sub>3</sub> produced				Mgs. N. as NH <sub>3</sub> produced				Mgs. N. as NH <sub>3</sub> produced			
	1.	2.	Avg.	% N. avail.	1.	2.	Avg.	% N. avail.	1.	2.	Avg.	% N. avail.
B. mesentericus .....	14.84	13.16	14.00	10.63	4.62	4.20	4.41	3.35	3.71	4.13	3.92	2.97
Ps. putida .....	27.72	24.78	26.25	19.94	5.04	5.04	5.04	3.83	4.41	4.97	4.69	3.56
B. vulgatus .....	21.14	15.82	18.48	14.04	4.48	3.92	4.20	3.19	4.69	3.71	4.20	3.19
B. megatherium .....	6.58	9.24	7.91	6.01	2.66	.....	2.66	2.02	3.29	.....	3.29	2.50
B. mycoides .....	15.54	14.42	14.98	11.38	1.96	2.38	2.17	1.64	5.25	5.67	5.46	4.14
B. subtilis .....	21.00	21.28	21.14	16.06	3.92	3.64	3.78	2.87	4.13	5.11	4.62	3.51
B. tumescens .....	13.86	16.80	15.33	11.64	3.36	3.50	3.43	2.60	10.57	14.21	12.39	9.41
S. lutea .....	28.00	27.02	27.51	20.90	5.04	2.52	3.78	2.87	3.71	4.69	4.20	3.19
B. proteus vulgaris....	17.08	13.30	15.19	11.54	9.38	8.82	9.10	6.91	5.11	3.43	4.27	3.24
B. ieteroides .....	8.40	lost	8.40	6.38	4.90	2.66	3.78	2.87	3.57	2.87	3.22	2.44
B. ramosus .....	7.98	9.10	8.54	6.48	6.30	5.18	5.72	4.34	5.53	3.85	4.69	3.56
Streptothrix, sp. ....	9.38	9.24	9.31	7.07	5.88	5.04	5.46	4.14	4.97	.....	4.97	3.77
Ps. fluorescens .....	8.40	7.84	8.12	6.17	.....	.....	.....	.....	.....	.....	.....	.....
B. vulgaris .....	29.68	34.44	32.06	24.36	7.28	7.70	7.49	5.69	2.03	2.45	2.24	1.70
Mic. tetragenus .....	22.68	21.42	22.05	16.75	6.02	6.58	6.30	4.79	6.37	7.63	7.00	5.31

tion. Of the three organisms showing the highest efficiency, as above indicated, in transforming the nitrogen of dried blood to ammonia, *B. vulgaris* appears in its turn to be the most vigorous, though the other two approach it closely and are about equal among themselves.

### *Clay-Loam Soil*

When the clay-loam soil is used as the medium with dried blood, marked differences are apparent in the efficiency of all the organisms. The poorer air supply of the clay-loam soil, due to its fineness and much greater tenacity, are evidently inimical to ammonia production, even though the same source of nitrogen—dried blood—is supplied for the ammonification process. The largest amount of ammonia produced in this series was that by *B. proteus vulgaris*, and even that was little more than one quarter of the amount produced by *B. vulgaris* as above noted in the sandy soil.

Moreover, the most efficient transformers of the nitrogen in dried blood in the sandy soil medium are not necessarily the same as those in the clay-loam soil. For example, in the case of the clay-loam soil *B. proteus vulgaris* is the most efficient ammonia producer with dried blood as ammonifiable material, while in the sandy soil the same organism manifests less than half the ammonifying efficiency of *B. vulgaris*. This latter organism, however, stands second to *B. proteus vulgaris* in efficiency in the clay-loam soil and yields 7.49 mgs. of ammonia nitrogen as against 9.10 mgs. produced by the last-named organism. There appear to be four other organisms which approach the efficiency of the two just discussed in the clay-loam soil in the following order: *Mic. tetragenus*, *B. ramosus*, *Streptothrix*, sp., and *Ps. putida*. While the latter does not compare in efficiency in the clay-loam soil with that shown by it in the sandy soil, it still manifests a notable efficiency. *Sarcina lutea*, however, appears to have lost in the clay-loam soil the marked ammonifying power possessed by it in the sandy soil.

Of course it must be again emphasized that the amounts of ammonia produced by all the organisms in the clay-loam soil

series are relatively so small that the margin allowable for safe comparison must of necessity be much decreased and therefore comparisons are more difficult.

#### CLAY-ADOBE SOIL

Passing on to a study of the data obtained with the clay-adobe soil as a medium, we find again that the physical condition of the soil is a powerful factor in determining the amount of ammonia produced in soils by pure cultures of organisms possessed of ammonifying powers, if the fifteen different organisms used are a suitable criterion. This confirms the findings of J. G. Lipman in his long series of ammonification experiments with mixed cultures. For practical purposes, we may add that most of the bacteria ammonify dried-blood nitrogen equally well in the clay-adobe soil and in clay-loam soil, though there does appear to be a slight though consistently greater amount of ammonia produced in the first-named soil. Again, we find in the clay-adobe soil an organism which stands out as far superior to all others in ammonifying efficiency and again also it is not the same organism as manifested that superiority in the preceding soil. While the duplicate determinations here do not agree as well as might be desired, they indicate amounts so much greater than the quantities of ammonia produced by the other organisms of the series that there can be no doubt of the marked and superior efficiency of *B. tumescens* as an ammonia producer from the nitrogen of dried blood in the clay adobe soil as a medium. Two other organisms appear to be in the second class in this series and they are *Mic. tetragenus* and *B. mycoides*. The first, it can be seen from Table III, occupied third place in efficiency in the clay loam soil, but the second has thus far been relatively inefficient. All the other organisms of the series do not manifest differences in efficiency of sufficient magnitude to warrant further comment, except that it is curious and interesting to note that the most efficient organism in the sandy soil is the least efficient organism in the adobe soil.

It should be remarked here that the generally excellent agreement between duplicate determinations, as shown in the table,

eliminates the fear that physical conditions in the individual cultures might operate to produce the rather marked effects noted.

In a comparison of the three soils it appears that the following organisms are among those which show the highest efficiency in transforming the nitrogen of dried blood into ammonia: *Ps. putida*, *Sarcina lutea*, *B. vulgaris*, *B. proteus vulgaris*, *Mic. tetragenus*, *B. tumescens*. Of these organisms only *Mic. tetragenus* shows a high efficiency in all three soils. Of the others *B. vulgaris* shows a high efficiency in both the sandy and adobe soils and *Ps. putida* in the sandy and clay-loam soil. The rest namely *Sarcina lutea*, *B. proteus vulgaris* and *B. tumescens*, are markedly efficient only in one soil each, namely, the sandy soil for the first, the clay-loam soil for the second, and the clay-adobe soil for the third. It appears to us particularly worthy of note, also, that of the last three organisms named *B. proteus vulgaris* and *B. tumescens* each holds a pre-eminent position of efficiency in its own soil which no other organism of the fifteen has approached; and even in the case of *Sarcina lutea* we find that it occupies a position in efficiency in its favorite soil which is second only to that of *B. vulgaris* and not very far behind the latter. This rather remarkable condition would hardly seem to be accidental and appears to us to indicate for certain organisms marked preferences for certain physical characteristics in media in which they are grown.

It must be added here, too, that only six of the fifteen organisms show marked ammonifying efficiency as regards the nitrogen of dried blood even when tested in three widely different soil types. The others vary but little from one another in all soils. Oddly enough, the organism which has shown the highest ammonifying efficiency because it maintained it through all soils, namely, *Mic. tetragenus*, has never before been looked upon, so far as we are aware, as an important ammonifier. That it should be the only one of fifteen organisms tested which should be about equally efficient in all soils used is not unworthy of note.

It is further significant to note here that *B. mycoides*, which has always been regarded as one of the most efficient soil organisms at ammonification, does not in our experiments show any unusual activity in that direction, at any rate when dried blood

is used as the ammonifiable material, no matter what the soil medium. A possible exception to this statement may be found in the case of the adobe soil, in which *B. mycoides* seems to be superior to all but three or four of the fifteen organisms tested. Such superiority, under the circumstances noted, is probably of little significance.

## SERIES II. EXPERIMENTS WITH TANKAGE

This series was arranged in a manner similar to the preceding, the sandy, clay loam, and clay-adobe soils again being used as media, but the ammonifiable material in this case was a high-grade tankage, the nitrogen content of which was 9.62 per cent. The results of the ammonia determinations were as shown in Table IV on page 150.

## DISCUSSION OF SERIES II

### *The Sandy Soil*

When we study in the foregoing table, the ammonification of tankage by pure cultures of bacteria, and compare the results with those of Table III, we see at once some very striking differences between the ability of the same micro-organisms to produce ammonia from tankage and from dried blood respectively. Not only do more of the organisms show a high efficiency in transforming the tankage nitrogen to ammonia, but the point of highest efficiency is not reached by the same organisms as before, others having taken their places in this series with sandy soil as the culture medium. For example, we find that *B. mesentericus*, which in the preceding series showed throughout an extremely low ammonifying efficiency even in the sandy soil, now manifests in the same culture medium, which, however, has tankage in place of blood added to it, the highest efficiency of all of the organisms tested. Indeed, it occupies a position of its own in that direction, much as does *B. vulgaris* in the sandy soil of the last series. The organism which approaches it most closely in the same medium is *B. proteus vulgaris* which, however, falls 25 per cent short of producing the amount of ammonia yielded by

TABLE IV  
AMMONIFICATION OF TANKAGE

Name of organism	SANDY SOIL				CLAY-LOAM SOIL				CLAY-ADobe SOIL			
	Mgs. N. as NH <sub>3</sub> produced				Mgs. N. as NH <sub>3</sub> produced				Mgs. N. as NH <sub>3</sub> produced			
	1.	2.	Avg.	% N. avail.	1.	2.	Avg.	% N. avail.	1.	2.	Avg.	% N. avail.
<i>B. mesentericus</i> .....	30.24	32.34	31.29	32.52	12.32	11.78	12.05	12.52	5.95	6.93	6.44	6.69
<i>Ps. putida</i> .....	17.50	15.96	16.73	17.39	8.00	8.96	8.48	8.81	6.23	6.65	6.44	6.69
<i>B. vulgatus</i> .....	20.02	16.24	18.13	18.84	12.60	11.50	12.05	12.52	7.63	7.07	7.35	7.64
<i>B. megatherium</i> .....	8.68	10.64	9.66	10.04	7.16	8.00	7.58	7.87	8.05	6.93	7.49	7.78
<i>B. mycoides</i> .....	14.94	13.02	13.93	14.48	11.06	11.50	11.28	11.72	9.59	11.27	10.43	10.84
<i>B. subtilis</i> .....	14.14	14.14	14.14	14.69	11.76	11.06	11.41	11.86	9.31	10.57	9.94	10.33
<i>B. tumescens</i> .....	22.26	21.10	21.98	22.84	12.90	11.62	12.26	12.74	10.57	12.25	11.41	11.86
<i>S. lutea</i> .....	15.96	.....	15.96	16.58	8.98	7.58	8.78	9.12	13.37	10.57	11.97	12.44
<i>B. proteus vulgaris</i> .....	22.96	26.04	24.50	25.46	3.36	3.92	3.64	3.78	4.27	.....	4.27	4.43
<i>B. icteroides</i> .....	8.54	.....	8.54	8.87	3.36	3.22	3.29	3.42	1.75	2.73	2.24	2.32
<i>B. ramosus</i> .....	19.74	20.72	20.23	21.02	12.46	7.30	9.88	10.27	5.67	6.37	6.02	6.25
<i>Streptothrix</i> , sp. ....	12.74	14.84	13.79	14.33	8.00	7.16	7.58	7.87	8.47	9.87	9.17	9.53
<i>Ps. fluorescens</i> .....	9.10	6.44	7.77	8.07	.....	.....	.....	.....	.....	.....	.....	.....
<i>B. vulgaris</i> .....	20.86	19.04	19.95	20.73	9.96	11.90	10.93	11.36	8.19	6.65	7.42	7.71
<i>Mic. tetragenus</i> .....	15.40	14.28	14.80	15.38	5.04	4.90	4.97	5.16	6.37	5.25	5.81	6.04



*B. mesentericus*. Then follow, not far behind *B. proteus vulgaris*, *B. tumescens*, *B. ramosus*, and *B. vulgaris* in the order named, the latter being more than  $33\frac{1}{3}$  per cent short of the efficiency exhibited by *B. mesentericus*. Nearly all of the other organisms fall more than 50 per cent short of the efficiency of the last-named organism under this set of circumstances, namely, sandy soil with tankage. There are, therefore, but five organisms out of the fifteen tested which can be adjudged distinctly efficient ammonifying organisms under these conditions. *B. mycoides* again exhibits a low efficiency, and not far different from, though slightly below, that manifested by it in the same soil in the dried-blood series.

It must be added here that, with the exception of *B. mesentericus*, the efficient organisms in this part of Series II have also shown more or less marked efficiency in the preceding series.

### *The Clay-Loam Soil*

Most of the organisms tested in the clay-loam soil seem to have found the latter a more congenial medium for ammonification with tankage than they did when dried blood was present. In this part of Series II, three of the organisms, namely, *B. mesentericus*, *B. vulgatus*, and *B. tumescens*, were not only the most efficient ammonifiers but also about equal in their ammonifying power. It appears, therefore, that so far as tankage is concerned *B. mesentericus* is an equally efficient ammonifier in the clay-loam and sandy soils. *B. tumescens* has previously established its pre-eminent position among the fifteen organisms as an ammonifier of dried-blood nitrogen in the adobe soil, but *B. vulgatus* enters here for the first time as a markedly efficient ammonifier.

Only slightly behind the three organisms just discussed in their ammonifying efficiency as regards tankage nitrogen in the clay-loam soil are, in the order named, *B. subtilis*, *B. mycoides*, and *B. vulgaris*. While the latter has manifested its high efficiency in other series above described, the first two organisms named for the first time in the work thus far described show marked ammonifying ability. Only three organisms in this part

of Series II have shown themselves to be really weak ammonifiers, and they are *B. proteus vulgaris*, *B. icteroides*, and *Mic. tetragenus*. The first and the third of these, it will be remembered, have given evidence of marked efficiency under other circumstances, but the second has thus far been throughout an organism of low efficiency. It is striking to note the much greater uniformity which exists in this portion of Series II in the ammonifying powers of four-fifths of the organisms tested than that which obtains in other parts of this series and of other series.

### *The Clay-Adobe Soil*

Here again we find the great uniformity in ammonifying power between the larger number of bacteria tested which is characteristic of the foregoing section of Series II. On the other hand, the clay-loam soil seems to have been a more congenial medium than the adobe soil for the ammonification of tankage nitrogen, for larger amounts of ammonia are produced in it by the same organisms in the same period of incubation.

*Sarcina lutea* shows the highest efficiency as an ammonifier in this part of the series, but is only slightly more efficient than *B. tumescens*. The organism taking third place is *B. mycoides* which, indeed, is not far behind the other two. Next in order of importance and still very efficient ammonifiers are *B. subtilis* and *Streptothrix*, sp. The other organisms are considerably weaker ammonifiers than those just mentioned and *B. icteroides* again proves to be distinctly the weakest. Both *Sarcina lutea* and *B. tumescens* have, as mentioned above under other circumstances, plainly evidenced their high efficiency as ammonifiers and *B. mycoides* and *B. subtilis* have taken similar positions with respect to tankage in the clay-loam soil. *Streptothrix*, sp., it will be remembered, has also shown a high ammonifying power before in the case of the clay-loam soil when dried blood was used.

Comparing the three soils in this series with the same ones in the preceding series, it appears quite clear that taking them by and large, ammonifying bacteria manifest a much higher efficiency with high-grade tankage than with dried blood under similar conditions. Likewise also, in Series II a small number of

organisms mostly identical with those in Series I seem to manifest a distinctly high efficiency which the much larger number of the balance do not in most cases even approach. It is worthy of remark, moreover, that *B. mycoides* attains or rather approaches in portions of Series II its position of prime importance which has thus far so generally been accorded it among ammonifying organisms.

### SERIES III. COTTONSEED MEAL

The cottonseed meal used in this series showed on analysis a nitrogen content equal to 5.5 per cent. The experiment in this series was otherwise conducted like those of Series I and II, 2 per cent of cottonseed meal being used, or 1 gram per 50 grams of soil. The results are shown in Table III.

Again the efficiency of the organisms tested is much greater in the sandy soil than in either the clay-loam or adobe soils.

#### *Sandy Soil*

In this part of Series III the organism of greatest ammonifying efficiency is *B. tumescens*. Those approaching it closely in efficiency are *B. vulgatus*, *Sarcina lutea*, and *B. mycoides* in the order named. *B. ramosus* takes fifth place and the next three organisms fall more than  $33\frac{1}{3}$  per cent short of producing the amount of ammonia yielded by *B. tumescens*. They are *B. mesentericus*, *B. megatherium*, and *B. proteus vulgaris*. The balance of the organisms show only about half the efficiency of *B. tumescens* and in one case, *B. icteroides*, which has in all previous series shown a very low efficiency, only about 25 per cent of the maximum efficiency is manifested. Taken as a whole, the data given in Table III for the Anaheim sandy soil reflects favorably on cottonseed meal as a source of available nitrogen under the conditions named. Again, the greatest efficiency is manifested in this section of Series III by organisms which in all cases have shown high efficiency in other series above reported. As in the case of the heavy soil in the preceding series with tankage, *B. mycoides* also shows marked efficiency in the light soil when cottonseed meal is used.

TABLE V  
AMMONIFICATION OF COTTONSEED MEAL

Name of organism	SANDY SOIL			CLAY-LOAM SOIL			CLAY-ADOBÉ SOIL		
	Mgs. N. as NH <sub>3</sub> produced			Mgs. N. as NH <sub>3</sub> produced			Mgs. N. as NH <sub>3</sub> produced		
	1.	2.	Avg. % N avail.	1.	2.	Avg. % N avail.	1.	2.	Avg. % N avail.
<i>B. mesentericus</i> .....	9.59	10.10	9.85	4.06	3.78	3.92	4.76	4.34	4.55
<i>Ps. putida</i> .....	8.75	7.07	7.91	3.22	3.50	3.36	4.06	4.34	4.20
<i>B. vulgatus</i> .....	12.39	11.83	12.11	22.02	4.90	5.39	4.20	4.88	4.54
<i>B. megatherium</i> .....	9.59	9.73	9.66	17.55	4.20	4.76	4.62	5.46	5.04
<i>B. mycoides</i> .....	11.13	11.55	11.34	20.61	2.52	2.87	7.28	5.04	6.16
<i>B. subtilis</i> .....	8.89	8.75	8.82	15.05	2.94	3.22	7.98	8.40	8.19
<i>B. tumescens</i> .....	12.67	14.07	13.37	24.30	5.04	3.78	8.82	11.20	10.01
<i>S. lutea</i> .....	11.69	12.25	11.97	21.75	2.38	1.96	4.34	5.46	4.90
<i>B. proteus vulgaris</i> .....	9.73	9.45	9.59	17.43	2.38	1.82	3.36	.....	3.36
<i>B. icteroides</i> .....	4.41	3.15	3.78	6.87	2.10	1.96	3.50	2.66	3.08
<i>B. ramosus</i> .....	10.15	10.57	10.36	18.84	7.28	6.72	7.84	5.88	6.86
<i>Streptothrix</i> , sp. ....	5.95	4.41	5.18	9.42	6.86	.....	4.90	4.06	4.48
<i>Ps. fluorescens</i> .....	8.33	.....	8.33	15.15	.....	.....	.....	.....	.....
<i>B. vulgaris</i> .....	7.07	8.75	7.91	14.37	6.30	3.50	3.92	3.78	3.85
<i>Mic. tetragenus</i> .....	9.17	6.51	7.84	14.25	1.54	1.96	2.24	3.92	3.08

### *The Clay-Loam Soil*

The most notable thing in this part of Series III is as above intimated, the very low efficiency of all of the organisms tested. Indeed only two organisms manifest any notable activity as ammonifiers of nitrogen in cottonseed meal in the clay-loam soil as a medium. These two in the order of their importance are *B. ramosus* and *Streptothrix*, sp. To these, in view of the disagreement of the duplicates as above shown, *B. vulgaris* may probably be added, and perhaps also *B. vulgatus*. The other organisms are all distinctly below the first two mentioned, and the lowest efficiency thus far noted is that exhibited by *Mic. tetragenus* in this soil. Considering the high efficiency of the latter organism in Series I, the results just discussed are puzzling.

### *The Clay-Adobe Soil*

Even a casual glance at the data obtained in the clay-adobe soil as a medium indicates the distinct superiority of that medium to the clay loam soil for ammonification of the nitrogen of cottonseed meal. Not only relatively but absolutely the data obtained show the production of much larger amounts of ammonia in this portion of the work.

As is the case in the sandy soil with cottonseed meal as the ammonifiable material, *B. tumescens* shows its distinct superiority to the other organisms as an ammonifier in the adobe soil. The next most efficient organism is *B. subtilis*, which, however, is considerably less efficient; and the next two organisms, about as far below *B. subtilis* in efficiency as the latter is below *B. tumescens*, are *B. ramosus* and *B. mycoides*. The other organisms are all low in efficiency, though in nearly all cases absolutely better than the same organisms in the clay-loam soil. Thus far we find that *B. subtilis* shows itself markedly efficient, for the first time, in the clay-adobe soil with cottonseed meal. All the other organisms above named have manifested marked efficiency in some parts of the foregoing series.

Looking at Series III as a whole it is interesting to note that the nitrogen of cottonseed meal seems to be made available through the activity of pure cultures of ammonifying bacteria,

with much greater rapidity than has heretofore been believed. The next striking fact brought out in these results is the clearly indicated superiority of the clay-adobe soil to the clay-loam soil as a medium for the ammonification of the nitrogen in cottonseed meal. The third point worthy of mention in Series III is the fact that we find again in it, as in the preceding series, only a few of the fifteen organisms tested which show marked ammonifying efficiency.

The culture of *Ps. fluorescens* died in the midst of these investigations and was not replaced.

#### SERIES IV. FISH GUANO

Fish guano, in accordance with the teachings of Voorhees and other agricultural chemists, has always been esteemed a good source of available nitrogen; in the words of Voorhees,<sup>2</sup> "ranking in availability well up to blood and tankage." It seemed to us therefore of importance to compare in these pure culture studies fish guano with the other organic materials discussed above. Accordingly a series was started similar to those above described, except that 1½ grams of finely sifted fish guano was the ammonifiable material used per 50 grams of soil. The fish guano used contained 8.63 per cent nitrogen. The results obtained are shown in Table VI.

The data in Table VI not only seem to confirm the opinion of Voorhees as above stated, if ammonia production by pure cultures may be taken as a criterion for determining the availability of fish guano, but they indicate in most striking fashion what was not shown in any of the foregoing series for the fertilizers, and particularly in the sandy soil, namely, the obliteration of the marked physiological differences obtaining in other series between the different organisms. Of the four nitrogenous fertilizers thus far discussed, fish guano seems to contain the form of nitrogen most generally ammonified by a large group of bacteria.

TABLE VI  
AMMONIFICATION OF FISH GUANO

Name of organism	SANDY SOIL				CLAY-LOAM SOIL				CLAY-ADONIS SOIL			
	Mgs. N. as NH <sub>3</sub> produced				Mgs. N. as NH <sub>3</sub> produced				Mgs. N. as NH <sub>3</sub> produced			
	1.	2.	Ave. % N. avail.		1.	2.	Ave. % N. avail.		1.	2.	Ave. % N. avail.	
<i>B. mesentericus</i> .....	10.64	.....	10.64	8.22	14.48	13.78	14.13	10.91	3.78	3.90	3.84	2.96
<i>Ps. putida</i> .....	15.68	13.86	14.77	11.41	10.00	10.98	10.49	8.10	3.50	3.90	3.70	2.85
<i>B. vulgatus</i> .....	14.28	14.56	14.42	11.14	9.58	11.54	10.56	8.15	2.36	3.50	2.93	2.26
<i>B. megatherium</i> .....	9.10	8.96	9.03	6.98	15.32	14.90	15.11	11.66	4.90	7.28	6.09	4.70
<i>B. mycoides</i> .....	10.08	9.38	9.73	7.51	4.68	4.12	4.40	3.39	9.10	7.56	8.33	6.43
<i>B. subtilis</i> .....	10.78	11.76	11.27	8.70	8.32	7.90	8.11	6.26	9.80	8.82	9.31	7.19
<i>B. tumescens</i> .....	16.38	16.80	16.59	12.82	9.30	10.98	10.14	7.83	8.12	12.46	10.58	8.17
<i>S. lutea</i> .....	13.16	14.28	13.72	10.60	8.74	7.06	7.90	6.10	3.08	5.60	4.34	3.35
<i>B. proteus vulgaris</i> .....	9.52	9.24	9.38	7.24	3.14	4.26	3.70	2.85	10.08	7.56	8.82	6.81
<i>B. icteroides</i> .....	12.04	13.72	12.88	9.95	4.82	4.70	4.76	3.67	0.84	0.00	0.42	0.32
<i>B. ramosus</i> .....	13.58	14.14	13.86	10.71	10.42	10.28	13.35	7.99	2.66	3.08	2.87	2.21
<i>Streptothrix</i> , sp. ....	8.96	7.98	8.47	6.54	4.82	4.40	4.61	3.55	5.60	7.84	6.72	5.18
<i>Ps. fluorescens</i> .....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
<i>B. vulgaris</i> .....	16.10	17.78	16.94	13.09	10.28	13.50	11.89	9.18	9.10	13.30	11.20	8.64
<i>Mic. tetragenus</i> .....	11.20	10.50	10.85	8.38	7.48	5.10	6.29	4.85	4.62	3.36	3.99	3.08

### *The Sandy Soil*

The good ammonifying power manifested by most of the organisms tested in this soil with fish guano finds its maximum in the case of *B. vulgaris*, which, it will be remembered, has already given evidence of notable ammonifying efficiency with other nitrogenous fertilizers. *B. tumescens*, likewise an organism with a record as an ammonifier well established, is a very close second to *B. vulgaris* in its efficiency at the ammonification of the nitrogen in fish guano. Four other organisms distinctly in the second class, and about alike in ammonifying efficiency in this part of Series IV, are, in the order of their efficiency, *Ps. putida*, *B. vulgatus*, *B. ramosus*, and *Sarcina lutea*. Peculiarly enough, *B. icteroides* shows an ammonifying efficiency here which places it in the second class, a position which it has thus far never even remotely approached in the series of experiments above discussed, nor, for that matter, in other series which are described below. All other organisms in this part of Series IV are distinctly in the third class, but nevertheless manifest notable ammonifying efficiency.

In no other series of results have we obtained such sharp lines of demarcation between the classes of organisms here tested, and arbitrarily grouped, in accordance with their respective powers of transforming organic nitrogen into ammonia.

### *The Clay-Loam Soil*

The change in the physical condition of the soil from the sand to the clay loam shows a marked effect on the ammonifying power of the same organisms. Nevertheless we find, on the whole, the best set of results thus far obtained with the clay loam when the series with fish guano is studied. In a class by itself under these conditions is *B. megatherium*, which is markedly superior to all other organisms in this part of Series IV, except *B. mesentericus*, which is a close second. The organism, however, which takes third place, *B. vulgaris*, falls about 20 per cent short of attaining the efficiency of *B. megatherium* under these conditions. Distinctly lower in efficiency in this part of Series IV are, in the order of their importance, but only slightly different from



one another, *B. vulgatus*, *Ps. putida*, *B. ramosus*, and *B. tumescens*. All of these, however, fall more than 30 per cent short of attaining the ammonifying efficiency of *B. megatherium* under the conditions here considered. Two other organisms, *B. subtilis* and *Sarcina lutea*, fall 40 per cent short of the efficiency of *B. megatherium*, and the others all fall far below even that figure.

It is singular, here again, that the physical nature of the soil medium employed should so strikingly and so variously influence the efficiency of the ammonifying bacteria. The two organisms which are distinctly superior in ammonifying ability as regards the nitrogen of fish guano in the clay loam soil were only of moderate efficiency with the same form of nitrogen in the sandy soil. And again, the organism which in the latter soil was paramount in its position retreats in the clay-loam soil to third place.

### *The Clay-Adobe Soil*

This is the first series of those we have thus far considered, as the data in Table VI shows, in which the clay-adobe soil proves, on the whole, to be inferior to the clay-loam soil. Seven organisms out of fourteen here show an extremely low ammonifying efficiency, and two others are by no means efficient organisms. That leaves five organisms in this group which may be considered of importance. Of these, *B. vulgaris* is the most efficient, but *B. tumescens* is not far behind it. *B. subtilis* belongs to the second class in this group and *B. proteus vulgaris* and *B. mycoides* to the third class. It is rather unfortunate that it was not possible in this part of Series IV to obtain better agreement between duplicate determinations. It would not appear to us, however, that the discrepancies in question militate against the justice of the conclusions above drawn.

*B. megatherium* holds a very good place in the sandy and clay loam soils of Series IV and a fair place in the adobe soil. The same is even more strikingly true of *B. vulgaris*, and in a minor degree this is also true of *B. tumescens*. The other organisms do not manifest such consistent efficiency under the three widely varying soil conditions.

## MISCELLANEOUS SERIES

For the purpose of comparing sheep and goat manure as well as phosphatic guano with the other organic materials above described and with peptone, it was deemed of interest to obtain data exemplifying the ammonification in the same soil of all the different materials above used with the three additional ones just mentioned. The sandy soil was chosen for this series and when peptone (Witte) was used, 0.5 gram of it was added to 50 grams of soil. The peptone contained 14.14 per cent N. The sheep and goat manure (2.13 per cent N), owing to its low nitrogen content, was added to the extent of 3 grams per 50 grams of soil, and the phosphatic guano (3.96 per cent N) was also added at the rate of 3 grams per 50 grams of soil. The results obtained are recorded in Table VII.

As was to be expected, the very available form of nitrogen in the peptone allows of the production of much larger quantities of ammonia than do the less available forms of the other materials. The fineness of division and easy solubility of the peptone, as well as the form of nitrogen which it contains, doubtless have contributed to the results. The more or less uniform decomposition of it, however, by most of the organisms tested shows peptone to be unsuited, as has been claimed by other investigators, for ammonification studies with pure or mixed cultures when the application of the data obtained, to field conditions, is contemplated. However that may be, *Sarcina lutca* shows the highest efficiency at ammonifying peptone nitrogen and at least six other organisms approach it rather closely. Relatively speaking, all but two of the organisms tested are efficient ammonifiers of peptone nitrogen. But their position with respect to peptone, as can be seen from Table VII, is no criterion as to their efficiency with respect to the other materials.

One of the interesting facts about the ammonification of the sheep and goat manure by pure cultures is that only one organism showed ammonifying efficiency worthy of the name, and that was *B. megatherium*. The other organisms showed a very slight power only of ammonifying the nitrogen in it. Just why this large discrepancy should exist with respect to this manure between *B. megatherium* and the other organisms still remains

TABLE VII  
AMMONIFICATION IN SANDY SOIL

Name of organism	BAT GUANO				SHEEP AND GOAT MANURE				PEPTONE			
	1.	2.	Ave.	% N avail	1.	2.	Ave.	% N avail	1.	2.	Ave.	% N avail
<i>b. mesentericus</i> .....	29.61	30.87	30.24	25.45	2.38	2.10	2.24	3.50	19.60	18.90	19.25	27.22
<i>b. putida</i> .....	32.15	33.11	32.62	27.45	3.08	3.36	3.22	5.03	20.16	20.02	20.09	28.41
<i>b. vulgaris</i> .....	37.45	34.79	36.12	30.40	2.52	4.62	3.57	5.58	13.30	15.68	14.49	20.49
<i>b. megatherium</i> .....	37.03	36.47	36.75	30.93	15.82	16.10	15.96	24.97	22.40	23.52	22.96	32.47
<i>b. mycoides</i> .....	45.14	40.53	42.84	36.06	2.38	2.62	2.52	3.95	26.32	.....	26.32	37.22
<i>b. subtilis</i> .....	34.23	32.55	33.39	28.10	1.40	1.96	1.68	2.63	27.16	28.28	27.72	39.20
<i>b. tumescens</i> .....	36.05	.....	36.05	30.34	2.38	2.24	2.31	3.61	25.06	24.78	24.92	35.24
<i>b. lutea</i> .....	25.27	25.83	25.55	21.50	2.10	2.24	2.17	3.39	30.10	29.26	29.68	41.98
<i>b. proteus vulgaris</i> .....	33.53	29.61	31.57	26.56	2.94	3.36	3.15	4.91	23.66	20.30	21.98	31.08
<i>b. heteroides</i> .....	22.75	22.47	22.61	19.03	2.66	1.82	2.24	3.50	24.08	26.46	25.27	35.74
<i>b. ramosus</i> .....	32.69	34.23	33.46	28.16	3.22	2.94	3.08	4.82	16.52	16.80	16.66	23.56
<i>treptothrix</i> , sp. ....	25.13	27.23	26.18	22.03	2.52	2.24	2.38	3.72	24.36	26.60	25.48	36.03
<i>s. fluorescens</i> .....	19.53	20.51	20.02	16.85	2.66	2.33	2.50	3.91	.....	20.72	20.72	29.30
<i>b. vulgaris</i> .....	40.39	35.77	38.08	32.05	2.24	1.82	2.03	3.17	27.58	27.02	27.30	38.61
<i>b. tetrageus</i> .....	36.33	34.23	25.28	29.69	2.80	2.38	2.59	4.05	26.46	28.98	27.72	39.20

to be explained. If, therefore, the readiness of the transformation of its nitrogen into ammonia by pure cultures of ammonifiers is to be taken as a criterion, sheep and goat manure must be adjudged to contain a relatively unavailable form of nitrogen.

The most amazing evidence portrayed in Table VII is the very high availability of the nitrogen of bat guano, which is a phosphatic guano. While without question the fact of the large amount of the guano used, as well as its low content of nitrogen, preclude an accurate and wholly justifiable comparison of it with the other nitrogenous materials, one cannot help being struck by the large transformation of its nitrogen into ammonia which nearly all of the organisms tested can accomplish. In many ways, the transformation of the nitrogen of bat guano into ammonia resembles that of the transformation of peptone nitrogen. The differences between the ammonifying powers of the different organisms are, however, unquestionably more marked in the case of the bat guano. In its efficiency as an ammonifier of bat guano nitrogen, *B. mycoides* appears for the first time in all the series studied to be distinctly superior to all the other organisms tested. *B. vulgaris* easily takes second place. Not far behind, however, and about equal in efficiency, are *B. megatherium*, *B. vulgatus*, *B. tumescens*, and *Mic. tetragenus*. In the third class are *B. ramosus*, *B. subtilis*, and *Ps. putida*, *B. proteus vulgaris*, and *B. mesentericus*. In the fourth class are *Streptothrix*, *sp.*, *Sarcina lutea*, *B. icteroides*, and *Ps. fluorescens*.

#### A COMPARISON OF THE RELATIVE AVAILABILITIES OF THE ORGANIC MATERIALS ABOVE EMPLOYED BASED ON THE PERCENTAGE OF NITROGEN CONTAINED IN THEM THAT WAS TRANSFORMED TO AMMONIA

Thus far we have been considering only the relative degrees of efficiency as ammonifiers of the different organisms among themselves as respecting a given organic form of nitrogen in a given soil. There is possible, however, a further very interesting study of the data above given as a basis. We refer to the percentage of nitrogen which is transformed in the different materials into ammonia so as to give them a relative rating as to availability as

TABLE VIII

SHOWING PER CENT OF NITROGEN TRANSFORMED TO AMMONIA WITH ALL FERTILIZERS IN ALL SOILS

Name of organism	SANDY SOIL.				CLAY-LOAM SOIL.				CLAY-ADobe SOIL.			
	Blood	Tankage	Cottonseed Meal	Fish Guano	Blood	Tankage	Cottonseed Meal	Fish Guano	Blood	Tankage	Cottonseed Meal	Fish Guano
<i>B. mesentericus</i> .....	10.63	32.52	17.91	8.22	3.35	12.52	7.12	10.91	2.97	6.89	8.27	2.96
<i>Ps. putida</i> .....	19.94	17.39	14.37	11.41	3.83	8.81	6.11	8.10	3.56	6.69	7.63	2.85
<i>B. vulgatus</i> .....	14.04	18.48	22.02	11.14	3.19	12.52	9.81	8.15	3.19	7.64	8.25	2.26
<i>B. megatherium</i> .....	6.01	10.04	17.55	6.98	2.02	7.87	8.14	11.66	2.50	7.78	9.16	4.70
<i>B. mycoides</i> .....	11.38	14.48	20.61	7.51	1.64	11.72	5.22	3.39	4.14	10.84	11.20	6.43
<i>B. subtilis</i> .....	16.06	14.69	15.05	8.70	2.87	11.86	5.61	6.26	3.51	10.33	14.89	7.19
<i>B. tumescens</i> .....	11.64	22.84	24.30	12.82	2.60	12.74	8.01	7.83	9.41	11.86	18.20	8.17
<i>S. lutea</i> .....	20.90	16.58	21.75	10.60	2.87	9.12	3.27	6.10	3.19	12.44	8.90	3.35
<i>B. proteus vulgaris</i> ...	11.54	25.46	17.43	7.24	6.91	3.78	3.30	2.85	3.24	4.43	6.11	6.81
<i>B. ieteroides</i> .....	6.38	8.87	6.87	9.95	2.87	3.42	3.69	3.67	2.44	2.32	5.60	0.32
<i>B. ramosus</i> .....	6.48	21.02	18.84	10.71	4.34	10.27	12.72	7.99	3.56	6.25	12.47	2.21
<i>Streptothrix</i> , sp. ....	7.07	14.33	9.42	6.54	4.14	7.87	12.84	3.55	3.77	9.53	8.14	5.18
<i>Ps. fluorescens</i> .....	6.17	8.07	15.15	.....	.....	.....	.....	.....	.....	.....	.....	.....
<i>B. vulgaris</i> .....	24.36	20.73	14.37	13.09	5.69	11.36	8.91	9.18	1.70	7.70	7.00	8.64
<i>Mic. tetragenus</i> .....	16.75	15.38	14.25	8.38	4.79	5.16	3.18	4.85	5.31	6.04	5.41	3.08

regards the work of pure cultures. The columns on availability of the various tables show a comparison in different soils of all the materials as attacked by the same organisms, and give the percentages of nitrogen in those materials which were transformed to ammonia. In Table VIII all of the columns indicating percentages of nitrogen of different fertilizers made available are brought together from the other tables and one is enabled to compare with much greater ease the different materials on the basis of availability in the same soil and with the same organism and in different soils, with different organisms.

### *The Sandy Soil*

So far as this soil is concerned, the data in Table VIII indicate clearly the superiority, from the point of view of the availability of its nitrogen, of cottonseed meal to the other organic nitrogenous fertilizers with which it is compared. Not only are the absolute amounts of ammonia produced in most cases larger from cottonseed meal nitrogen than from other forms, but there are more organisms of the fifteen tested which can vigorously ammonify this form of nitrogen. So that to illustrate, there are but five organisms which have shown the power to transform 15 per cent or more of the nitrogen of dried blood into ammonia in the sandy soil. Under similar conditions there are ten organisms which hold such a record for cottonseed meal. Tankage shows itself to be the next important nitrogenous fertilizer to cottonseed meal from the point of view of the availability of its nitrogen. Thus, comparing it with dried blood as above, we find that there are nine organisms which transform 15 per cent or more of the nitrogen of tankage into ammonia. No such high availability is obtained at all in the case of the fish guano.

When we consider these nitrogenous fertilizers from the point of view of the transformation of 10 per cent or more of their nitrogen into ammonia, we find that there are thirteen of the fifteen organisms tested which possess that power as regards cottonseed meal nitrogen and another comes very close to that point. In the case of tankage nitrogen there are thirteen organisms with a similar power. In the case of dried-blood nitrogen

there are but ten organisms which can accomplish that task, and but seven such in the case of fish-guano nitrogen.

From the point of view of availability by pure cultures therefore in the sandy soil the four nitrogenous fertilizers are to be rated as follows: cottonseed meal, tankage, dried blood, and fish guano. The first two are nearly alike and are far superior to the last two, which are nearly alike, but much more different from each other than the first two.

The greatest efficiency at ammonification manifested by any organism in the sandy soil is that of *B. mesentericus* with tankage nitrogen, which transforms 32.52 per cent of the nitrogen present into ammonia in twelve days. It should be noted in this connection also that absolutely higher amounts of ammonia are produced from tankage nitrogen than from any other form in the sandy soil, even if there are fewer organisms which attack it readily than there are in the case of the cottonseed meal.

### *The Clay-Loam Soil*

Conditions are evidently entirely different for ammonification in this soil. Not only is the ammonia production very low so far as all the fertilizers are concerned, but they no longer bear to one another the relation which obtained between them in the sandy soil. There are thus but few organisms which possess the power of transforming 10 per cent or more of the total nitrogen in any of the four fertilizers into ammonia in twelve days. In fact, there are none such in the case of the dried blood, only two such each in the cases of cottonseed meal and fish guano, and seven such in the case of the tankage. No organism attains to the production of ammonia equivalent to 13 per cent of the total amount present in the clay-loam soil regardless of the kind of fertilizer at its disposal.

The tankage, however, is superior to the cottonseed meal in the clay loam and distinctly so as above indicated. The cottonseed meal takes second place, the fish guano third place, and the dried blood is by far the poorest. Indeed, no organism was capable of producing an amount of ammonia in excess of 6.91

per cent of the total amount of dried-blood nitrogen furnished, and that occurred in only one case, all the other organisms producing much less.

### *The Clay-Adobe Soil*

Some very striking facts become apparent when the availability of the four fertilizers in clay-adobe soil are considered. While on the whole dried-blood nitrogen is only slightly more efficiently transformed into ammonia than in the clay-loam soil, cottonseed meal and tankage, particularly the former, are more vigorously acted on in the clay-adobe soil by most of the organisms. Fish guano, while not markedly so, is none the less superior here again to dried blood.

Again comparing the different fertilizers on the basis of the amounts of their nitrogen transformed into ammonia by the organisms tested, we find that four organisms transform 10 per cent or more of the nitrogen in cottonseed meal into ammonia, a like number accomplish similar results in the case of tankage, and none succeeds in that direction in either fish guano or dried blood. While thus cottonseed meal and tankage appear alike, a study of table VIII reveals the superiority of the former in the larger absolute amounts of nitrogen which are transformed there than in the case of the latter.

The first striking fact shown in Table VIII is the marked superiority of the sandy soil as a medium for ammonification by pure cultures. Likewise the added fact of its superiority as a medium for most of the organisms tested must be noted in this connection. The second point of great interest is the surprising fact of the superiority of the clay-adobe soil to the clay-loam soil as a medium for ammonification. From its tenacious nature one would suppose the former type to be a much poorer medium for ammonification than the clay loam and yet it is distinctly superior to the latter as regards tankage and cottonseed meal. As regards fish guano, it is slightly inferior to the clay loam and again as regards the dried blood about equal to the clay loam or possibly slightly superior.

Considering all the data given in Table VIII from all points of view, tankage must be given first place as regards the avail-



ability of its nitrogen, cottonseed meal easily takes second place and, owing to its superiority in the sandy soil, dried blood takes third place. Considering the superiority of the fish guano to the dried blood in the other soils, however, it is probably fairer to adjudge dried blood and fish guano of equal availability from the point of view of the transformation of their nitrogen into ammonia by pure cultures of ammonifying bacteria.

### GENERAL DISCUSSION

Several of the facts which have come to light in the investigations above described demand a word of comment with respect to their general significance. First as regards the relative efficiencies at ammonification of the different organisms tested, we find that there is marked variation. Indeed it is difficult to find an organism among the fifteen tested which consistently stands as the best ammonifier regardless of the soil and the ammonifiable material employed. There are, however, one or two organisms which nearly approach such a description. In other words, it appears that, viewing ammonification of organic nitrogen from the standpoint of pure cultures, every organism will do best with a definite combination of soil and organic matter. To be sure there are some organisms of those tested, even though they be in the minority, which are consistently weak ammonifiers. *B. icteroides* and *Ps. fluorescens* serve to exemplify such.

That *B. mycoides* is by no means always the most efficient of ammonifying bacteria as has heretofore been believed is clearly indicated above. On the other hand, it does possess and manifest marked superiority in certain cases. Thus, for example, while showing poor or mediocre ammonifying power in different soils with dried blood, tankage and fish guano, it manifests great vigor in the case of cottonseed meal and succeeds in making the record for the percentage of nitrogen transformed in the case of bat guano, in which it transforms to ammonia 36.06 per cent of the nitrogen present.

Comparing the organisms in any one given soil as a medium we find some interesting facts. In the sandy soil, for example, with dried blood *B. vulgaris* is the most efficient ammonifier, making available or transforming into ammonia 24.36 per cent of the nitrogen present. With tankage, *B. mesentericus* shows the highest efficiency, transforming 32.52 per cent of the nitrogen present into ammonia. With cottonseed meal, *B. tumescens* is paramount, yielding an amount of ammonia equivalent to 24.30 per cent of the amount of nitrogen present. With fish guano, *B. vulgaris* again manifests its superiority over the other organisms by changing 13.09 per cent of the nitrogen present into ammonia. When bat guano is used, *B. mycoides* stands distinctly superior to all others, as above shown, by transforming 36.06 per cent of the nitrogen present into ammonia. In the case of sheep and goat manure there is but one efficient organism and that is *B. megatherium*, which transforms 24.97 per cent of the nitrogen present into ammonia. Lastly when peptone is used *Sarcina lutea* stands pre-eminent, and when all materials are compared, regardless of whether they were used in all soils or not, the last-named organism makes the record for availability by transforming 41.98 per cent of the nitrogen present into ammonia. There are thus six organisms out of the fifteen which make records in one and the same soil but with different forms of organic matter. One of the six stands superior in the cases of two nitrogenous materials, namely dried blood and fish guano, and that is *B. vulgaris*.

Comparing the same organisms with the same nitrogenous materials, above used, except the last three named, in the clay-loam soil, we find that with dried blood *B. proteus vulgaris* is most efficient, transforming 6.9 per cent of the nitrogen present into ammonia. With tankage, *B. tumescens* is pre-eminent and transforms 12.74 per cent of the nitrogen present into ammonia. With cottonseed meal, *Streptothrix*, sp., is superior, transforming 12.84 per cent of the nitrogen present into ammonia; and lastly with fish guano, *B. megatherium* is again pre-eminent, transforming 11.66 per cent of the nitrogen present into ammonia. We see again, therefore, that in one and the same soil, with four different materials, four different organisms make records for

availability. To be sure, two of these organisms have shown themselves superior to all others in the sandy soil but with different materials.

In the case of the clay-adobe soil we find that with dried blood *B. tumescens* stands at the top of the list and transforms 9.41 per cent of the nitrogen present into ammonia. With tankage in the same soil, *Sarcina lutea* is the most efficient ammonifier and transforms 12.44 per cent of the nitrogen present into ammonia. With cottonseed meal, *B. tumescens* is again superior to all others and transforms 18.20 per cent of the nitrogen present into ammonia; and finally with fish guano *B. vulgaris* again assumes the ascendancy and transforms 8.64 per cent of the nitrogen present into ammonia. *B. tumescens* is very nearly as efficient in this latter case as *B. vulgaris*. We have thus seen that organisms which in all cases have shown their superiority in other soils are also very efficient in the clay-adobe soil.

It is clear, therefore, that only about half of the fifteen organisms tested show greatest efficiency in some soil or with some form of organic matter. Scrutinizing more closely the efficiencies of each of these, we must concede to *B. tumescens* the paramount place among them, for it has stood pre-eminent, in five combinations of soil and fertilizer, out of fifteen, and has in addition been close to first place in several other instances.

Comparing our findings with those of Marchal, the following critical statements must be made. First, that results of solution cultures are no criterion as to results to be obtained in soils. Secondly, that no two forms of organic nitrogen are attacked and ammonified with the same vigor by any one organism. Thirdly, that different soils will modify an organism's power to ammonify any one given form of nitrogen very markedly, so that it may be efficient in one case and feeble in another. Fourthly, that the ammonifying efficiency of organisms is greater in sandy soil, and possibly in others, than in solutions, for we have obtained a transformation of 41.98 per cent of peptone nitrogen and 36.06 per cent of bat guano nitrogen into ammonia by *Sarcina lutea* and *B. mycoides* respectively in twelve days at temperatures between 27° C and 30° C, while Marchal only obtained similar transformations in thirty days at 30° C in albumin solutions.

Owing to the general distribution of the efficient ammonifying organisms above described in most soils, it is extremely improbable, to judge from our results, that we may look forward to a profitable form of inoculation of soils with ammonifying bacteria. The choice, however, of the form of nitrogenous fertilizer for a soil, which will be most readily made available, when available nitrogen is needed, may indeed be something of much greater practical significance, as our data would appear to indicate. Particularly emphatic are our results in illustrating that, at least so far as pure cultures are concerned, and as also partly shown by J. G. Lipman<sup>3</sup> and others, with mixed cultures, prevalent ideas with respect to the relative availability of common nitrogenous fertilizers are incorrect. Both tankage and cottonseed meal, and, in some cases, fish guano, show higher availabilities than dried blood, which we have always been in the habit of regarding as the most available of organic nitrogenous fertilizers (based only on vegetation tests).

Nor do we argue too far afield from our subject when we make the remarks just preceding. We recognize fully that availability as measured by ammonification does not necessarily denote availability as measured by nitrification nor by assimilation of nitrogen by plants. We cannot, however, help noting, also, that a persistent preference exists among practical orchardists in this state for tankage as against dried blood and we therefore feel that, judged by other criteria, similar conclusions, must be drawn to those adduced from our experiments with pure cultures of ammonifying bacteria.

We have decided, however, to go much further in these experiments and are now prosecuting more elaborate investigations on nitrogen transformation not only to ammonia but also to nitrates. In these studies we shall deal with the soil flora as existing in a large variety of soil types freshly collected from the field for our purposes. Moreover, we shall employ the raw, unsifted fertilizer material just as it is used by the farmer. From these investigations we hope to glean much more evidence which will be of great practical significance, and, like the results above described, also of marked scientific interest.

## SUMMARY

Results are above given which deal with:

1. The marked differences in ammonifying efficiency of fifteen organisms in pure culture.

2. The soil was used as a medium and three types employed, sandy, clay loam and clay adobe.

3. Four common fertilizers as sources of nitrogen were used, in all soils, and peptone, bat guano, and sheep and goat manure employed only in the sandy soil, besides. The four fertilizers were dried blood, tankage, cottonseed meal and fish guano.

4. The nature of the soil, as well as the nature of the nitrogenous material, markedly modify an organism's ammonifying power.

5. There is no regularity in these variations and they cannot be foretold.

6. While it is difficult to make an exact decision, *B. tumescens* appears, on the whole, to have been the most efficient organism above tested.

7. The highest efficiency in a single culture with a fertilizer was shown by *B. mycoides*, which transformed in twelve days at 27° to 30° C, 36.06 per cent of the nitrogen in bat guano into ammonia.

8. The highest efficiency in a single culture with peptone was shown by *Sarcina lutea*, which transformed 41.98 per cent of the nitrogen present into ammonia under similar conditions.

9. A comparison of availability of nitrogenous fertilizers with ammonifiability as a criterion, according to our experiments, markedly changes the positions of tankage, fish guano, and cottonseed meal with respect to dried blood, showing them in most or in many cases to be superior to the latter.

10. One half of the number of organisms tested is far superior to the other half in ammonifying ability.

11. Interesting comparisons with Marchal's work are made.

12. *B. icteroides* has shown itself throughout to possess but little ammonifying efficiency.

13. Experiments are following those above described to make the latter more complete. Elaborate tests are being made with many soil types using the original soil's mixed flora from freshly collected samples in the field. Not only ammonifying powers of these mixed flora in the different soil types will be studied, but also the corresponding nitrifying powers. A large variety of nitrogenous fertilizers as employed in practice will be tested here.

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- <sup>7</sup> *Ibid.*, vol. 15, p. 433.
- <sup>8</sup> *Ibid.*, vol. 29, p. 238.
- <sup>9</sup> *Ibid.*, vol. 31, p. 49.

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HUMUS AND HUMUS-NITROGEN IN  
CALIFORNIA SOIL COLUMNS

BY  
R. H. LOUGHRIDGE

UNIVERSITY OF CALIFORNIA PRESS  
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# HUMUS AND HUMUS-NITROGEN IN CALIFORNIA SOIL COLUMNS

BY

R. H. LOUGHRIDGE

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## INTRODUCTION

Humus, as distinct from the unhumified organic matter, is now regarded as one of the most valuable ingredients of fertile soils because of its physical effect and because of its content of nitrogen, potash, and phosphoric acid, which while not soluble in water, are rendered available to plant use by the action of soil bacteria. It has no definite chemical composition, for it is produced by the decay of vegetable and animal matters of all kinds in the soil and under certain conditions of warmth and moisture, and is naturally made up of the elements found in those substances; during the process of humification, however, there is a gain in the percentage of carbon and nitrogen because of corresponding losses of oxygen and hydrogen. The proportion of these and of mineral matters in the humus varies then with the nature of the substances from which they are derived, and probably with the degree of intensity of the action of each factor in the humification process, in which oxidation because of too great heat, or other changes because of excess of water may be brought about.

The amount of humus produced from organic matter depends upon the nature and condition of the materials used, and upon its complete humification by the maintenance of proper conditions in the soil. Professor Hilgard<sup>1</sup> thinks that "in the humid region one part of normal soil humus might be formed from five or six parts of dry plant debris; while in the extreme regime of the arid regions from eighteen to twenty parts of the same

<sup>1</sup> Soils (Macmillan & Co., 1906), p. 128.

would be required.” The experiments of Professor Harry Snyder of Minnesota<sup>2</sup> on the humification of different materials gave interesting results. Humification under favorable conditions was allowed to proceed a year in each case, at the end of which time the amount of humus obtained from each substance and its composition was ascertained. Oat straw and sawdust seemed unchanged even when humification was allowed to continue longer than a year. The following table illustrates some of Snyder’s findings:

TABLE 1.—HUMUS OBTAINED FROM DIFFERENT MATERIALS

Fresh cow manure	33 parts for 1 part of humus	6.16% nitrogen in humus
Green clover .....	25 parts for 1 part of humus	8.24% nitrogen in humus
Meat scraps .....	11 parts for 1 part of humus	10.96% nitrogen in humus
Sawdust .....	10 parts for 1 part of humus	0.30% nitrogen in humus
Oat straw .....	6 parts for 1 part of humus	2.50% nitrogen in humus

With ordinary green-manure crops and under field conditions it will probably require a much larger amount of green material to produce one part of humus, for there are losses from the incomplete turning-under of the material and the drying-out of the soil.

It would naturally be supposed that as humus is formed by the decay of roots, leaves, etc., it would be found only where formed, whereas on the contrary, we find it quite evenly diffused through the surface soil; this even distribution is not easily accounted for without some outside agencies. Professor Hilgard, in discussing the subject,<sup>3</sup> attributes it chiefly to the action of fungi, insects, and earthworms.

The vegetative fibrils (mycelia) of several kinds of molds are constantly present in the soil, and while consuming dead tissue of the higher plants, spread their own substance throughout the soil mass. . . . All these being dependent upon the presence of air for their life functions, remain within such distance from the surface as will afford adequate aeration; the depth reached depending upon the perviousness of the soil and subsoil. In the humid region this will usually be within a foot of the surface, but in the arid may reach to several feet. . . . The earthworm nourishes itself by swallowing, successively, portions of the surrounding earth, digesting a part of its organic matter, and ejecting the undigested earth in the form of “casts” such as may be seen by thousands on the surface of the

<sup>2</sup> Bulletin 53 Minn. Agr. Exp. Station.

<sup>3</sup> Soils (Macmillan & Co., 1906), p. 157.

ground during and after a rain. . . . In humid climates and in a ground fairly stocked with these worms the soil thus brought up may amount to from one-tenth to two-tenths of an inch annually over the entire surface; so that in half a century the entire surface foot might have been thus worked over. Aside from the mechanical effect thus achieved in loosening the soil, and the access of air and water permitted by their burrows, the chemical effects resulting from their digestive processes, and the final return of their own substance to the soil mass; also their habit of drawing after themselves into their burrows, leafstalks, blades of grass, and other vegetable remains, renders their work of no mean importance both from the physical and chemical point of view. . . . The work of earthworms is especially effective in loamy soils and in the humid regions. In the arid region and in sandy soils generally the life conditions are unfavorable to the worm, and the perviousness elsewhere brought about by its labors already exists naturally in most cases.

The amount and nature of humus depends much upon climatic factors as is especially noted in arid and humid regions. In the humid region with its frequent rainfall and a comparatively shallow soil, the vegetable material (roots, leaves, etc.) is held near the surface and to this depth the humus is limited; while in the very deep and warm soils of the arid region the penetration of plant roots is to depths of fifteen to thirty feet, and as a consequence of their decay humus is found to depths of twelve or fifteen feet and in some cases much deeper.

It is this deep distribution of humus throughout the agricultural regions of California that is treated of in detail in this bulletin, showing a fundamental difference between the soils of humid and arid regions.

## HUMUS IN CALIFORNIA SOILS

The study of the soils of California was begun by Professor Hilgard immediately after entering upon his duties in 1874 as Professor of Agriculture in the University of California, and his first report, made in 1877, contains physical and chemical analyses together with descriptions of a number of soils, as well as the results of alkali investigations. The analysis of a soil for its humus content is first given in the report of the Experiment Station for 1879, and since then nearly every annual report to and inclusive of 1904 contains soil descriptions and analyses, the percentage of humus being given for the first foot or sometimes

for the upper two feet, as it was not then known that the humus reached to a greater depth than three feet. In the report for 1904 the first analysis is given of a soil with humus to a depth of twelve feet.

During these years the call from farmers for information regarding the needs of their soils was very great, and thousands of soils were sent in for examination. These did not require an accurate and full analysis in every case, and were subjected to cursory tests<sup>4</sup> by which approximate determinations could be reached very quickly for phosphoric acid, lime, and humus. The results were graded into "poor" for less than 0.7 per cent; "fair" for 0.7 to 1.0 per cent; and "good" when the percentage was above 1.0, and only the surface soils were examined. From 1893 to 1908 there were made humus tests on nearly fifteen hundred soils distributed among the following agricultural regions; in this table are given the percentages of soils whose humus belonged to the several grades.

TABLE 2.—HUMUS IN SOILS OF THE AGRICULTURAL REGIONS; QUICK METHOD

Regions	No. of soils examined for humus	Percentage of soils having		
		Good humus	Fair humus	Poor humus
Sacramento Valley .....	211	55	18	27
San Joaquin Valley .....	434	24	24	52
Foothills of Sierra Nevada .....	66	59	12	29
Coast Range valleys .....	398	64	19	17
Southern California .....	347	43	23	34
Average .....	1,456	46	21	33

It will be seen from the above that more than one-half of the soils of the Sacramento, Foothill, and Coast Range regions that were sent for examination by farmers of those regions had as much as 1 per cent of humus, though for the state at large only 46 per cent of the 1456 soils had that amount.

<sup>4</sup> The method of this cursory examination for humus is as follows: Take two grams of air-dried, well-mixed soil of twelve inches depth, place in a test-tube of one-half inch diameter and add about ten cubic centimeters of 10 per cent solution of potassic hydrate. Boil for a few minutes and allow the soil to settle. The potash dissolves the humus and produces a dark color of greater or less intensity according to the amount of humus present. By taking soils in which the humus percentages have already been ascertained correctly, and treating them by this short method, a scale of color intensities may be adopted by which approximate percentage of humus may be reached in most of the soils.

The results of these soil examinations have brought out the fact that, while the supply of *potash* is as a rule very large and should not need replenishment by fertilizers for decades of years and that *phosphoric acid* is generally fair in amount, except in lands that have been in wheat culture for many years, the percentage of *humus* in all surface soils except tule swamps is quite low and has required a system of green manuring to bring the land back into the best condition.

*Regular Analyses.*—Since the establishment of the station, nearly six hundred complete analyses have been made of soils chosen to represent the different agricultural regions and conditions in the state, and among the determinations was that of the exact amount of humus in 331 surface soils. The method of analysis used was what is known as the Grandeau Method as modified by Professor Hilgard. The results, therefore, more nearly represent actual average conditions in the soils of the state. The following table shows the average percentages of humus in the soils of the several regions:

TABLE 3.—PERCENTAGES OF HUMUS IN SURFACE SOILS; REGULAR METHOD

Regions	No. of soils examined for humus	Average percentage of humus	Percentage of soils having more than 1% of humus
Tules and meadows .....	14	3.62	all
Coast Range valleys .....	91	1.69	67
Sierra Foothills .....	46	1.23	41
Sacramento Valley .....	29	1.14	50
Southern California .....	61	.94	28
San Joaquin Valley .....	67	.79	29
Desert plains and lava bed valleys	23	.51	0
General average for state .....	331	1.25	40

The general average of humus in the 331 surface soils taken from different parts of California and supposed to represent fairly all of the agricultural regions is 1.25 per cent. If, however, the tule marshes and the meadow lands are omitted from the calculation, then we find that the general humus percentage is 1.15.

DISTRIBUTION OF HUMUS DOWNWARD IN  
CALIFORNIA SOILS

A very marked characteristic of the soils of California is their *great depth*, as exemplified in the uniformity of color through a depth of several feet, their *good texture*, affording easy penetration of plant roots often to depths of twenty and even sixty feet below the surface, *abundant food* available for plant use throughout these depths, an *absence of any well-defined subsoil* except at a depth of several feet below the surface, the *absence of any compact clay substratum* differing from the surface foot, and the *presence of humus* to a depth of as much as ten or more feet. In each of these particulars the soils of California differ from those of the humid states east of the Rocky Mountains, and because of this the methods of culture are different, and in the applications of phosphate fertilizers to the soil it is only with great difficulty that they can be made to reach the feeding roots of plants.

There are several striking facts regarding the humus itself in the soils of California which deserve mention and which distinguish them from the soils of the humid region of the eastern states, one of which has but recently been brought out in the examinations that are being made of the one hundred and more typical twelve-foot columns from the chief agricultural divisions of the state. These facts are: *first*, the small percentage of humus in the first foot of all California soils as compared with the much higher percentages found in eastern soils; *second*, the distribution of the humus downward to depths of twelve and more feet in upland soils, the total amount being usually greater than that of the eastern soils; and *third*, the rather higher percentage of humus-nitrogen in the upper three feet, and the distribution of the humus-nitrogen throughout the twelve feet, thus giving the soil in the arid region a higher total of humus-nitrogen than is found in the humid soils.

*Bacterial Activity in the Soil.*—Another highly important advantage possessed by California in her soils is the fact recently brought out by Professor C. B. Lipman of this station in his paper, "The Distribution and Activities of Bacteria in Soils of the Arid Region":<sup>5</sup>

<sup>5</sup> Univ. Calif. Publ. Agr. Sci., vol. 1, no. 1, pp. 17, 20.

. . . that soils of the arid region at all depths studied show ammonifying powers, which are, however, generally most vigorous in the first six or eight feet. In one case ammonification was noted in soil from a depth of fifteen feet, or adjoining the water-table. . . . As for nitrification my data present again features of striking interest. They go to prove that nitrate formation, like ammonification, goes on at much greater depths in soils of the arid than in soils of the humid region. . . . That nitrification is found commonly down to a depth of five or six feet in soils of the arid region. In one case soil from an eight-foot depth showed a vigorous nitrifying power.

Professor Lipman's observations greatly emphasize the importance of having the roots of the various crops utilize the food-supplies at their command at depths of many feet; for not only is there a nitrogen supply, but our investigations on the soil columns show that the amount of available phosphoric acid and of potash is large at depths of twelve and more feet.

## DISTRIBUTION OF HUMUS AND HUMUS-NITROGEN IN SOIL COLUMNS FROM EACH AGRICULTURAL REGION

The agricultural regions of the state comprise the Sacramento and San Joaquin valleys, forming together the central Great Valley, the foothills lying on the western slope of the Sierra Nevada, the many Coast Range valleys and low hills among the mountain ranges along the western side of the state, the Southern California valleys and the low hills, the desert plain, which is being brought under cultivation by the development and use of irrigation water, and the northeastern lava-bed valleys.

As the object of this investigation was chiefly to ascertain the extent to which humus was found in the lower depths of the soils, the soil columns were taken only from those regions whose soils are many feet in depth. The number of columns obtained was 110 from thirty-seven counties, each column intended to represent a characteristic type of land in its particular region. There are, of course, very many small valleys and minor soil regions that have not been included in this investigation. The columns have been placed in vertical frames against the walls of the lecture room of Budd Hall at the University of California, arranged and labelled according to the respective agricultural regions. Each soil occupies an eight-ounce bottle and the



columns are nearly eight feet in height, thus forming a very conspicuous and interesting exhibit, probably the first and only one of its kind anywhere.

The percentage of humus and of humus-nitrogen has been ascertained in all of these soils as shown in the following pages of this bulletin. The chemical analysis of the first, sixth, and lowest foot of each column and the physical analysis of the first or upper foot have been nearly completed and the results, with proper discussion, will be given in future bulletins. The humus and nitrogen determinations in the bulletin were made chiefly by Messrs. M. E. Holter and F. H. Wilson, and the late F. E. Johnson, assistants in the soil laboratory.

#### SOIL COLUMNS OF THE SACRAMENTO VALLEY

The valley of the Sacramento River, lying between the two great mountain ranges—the Sierra Nevada on the east and the Coast Range on the west—which unite on the north, covers an area of 6200 square miles. The valley is widest on the south, where it unites with the San Joaquin Valley. Within its area are four or five general and highly distinct soil regions, or types, each of which is represented by one or more soil columns in our collection, or eighteen in all.

*Alluvial Lands.*—These border the Sacramento River and are timbered with sycamore, white oak, and ash. The soil is a dark loam with little or no change to the depth of twelve feet, as shown in the soil columns, and even deeper. Three columns were taken from the following localities: Near Cottonwood, Shasta County, by W. S. Guilford; Glenn Post Office, in Glenn County, east of Willows; and near Perkins, Sacramento County, by Professor George Roberts, now of the Kentucky Agricultural College. To these is added a column of the alluvial of Chico Creek taken from the Bidwell place, at Chico, Butte County.

The alluvial lands, with the exception of the sample from the hop fields at Perkins, may be regarded as rich in humus in the upper three feet, while all show fair amounts below that depth. The soil texture is good and it is to the advantage of plant roots that the humus with its nitrogen should be thus distributed, the sum in each of the four columns being from 5.60 to 8.80 per cent.

TABLE 4.—HUMUS IN COLUMNS OF ALLUVIAL LANDS OF SACRAMENTO VALLEY

SACRAMENTO RIVER										CHICO CREEK					
SHASTA COUNTY				GLENN COUNTY				SACRAMENTO COUNTY				BUTTE COUNTY			
COTTONWOOD				GLENN P. O.				PERKINS				CHICO			
Ft.	Soil Clay 12.54	Humus- Nitrogen in		Soil Clay 29.54	Humus- Nitrogen in		Soil Clay 4.49	Humus- Nitrogen in		Soil Clay 11.46	Humus- Nitrogen in				
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil			
1	Dark loam....	1.50	4.93 .07	Dark loam....	1.88	3.88 .07	Sandy .....	.66	7.21 .05	Sandy .....	1.84	6.03 .11			
2	Dark loam....	.91	8.13 .07	Dark loam....	1.50	3.47 .05	Sandy .....	.76	5.39 .04	Sandy .....	1.48	6.62 .10			
3	Dark loam....	.51	7.65 .04	Dark loam....	1.16	3.63 .04	Sandy .....	.78	5.74 .05	Sandy .....	1.16	6.90 .08			
4	Dark loam....	.60	9.83 .06	Dark loam....	.88	3.18 .03	Sandy .....	.76	5.76 .04	Sandy .....	.88	8.86 .08			
5	Dark loam....	.49	10.80 .05	Dark loam....	.72	4.30 .03	Sandy .....	.76	2.21 .02	Sandy .....	.58	8.27 .05			
6	Dark loam....	.53	8.49 .05	Dark loam....	.60	3.67 .02	Sandy .....	.64	2.63 .02	Sandy .....	.42	10.47 .04			
7	Dark loam....	.64	8.75 .06	Dark loam....	.40	4.26 .02	Sandy .....	.56	4.12 .02	Sandy .....	.40	13.00 .05			
8	Dark loam....	.52	7.50 .04	Dark loam....	.33	2.43 .01	Sandy .....	.34	6.18 .02	Sandy .....	.36	10.83 .04			
9	Dark loam....	.38	7.63 .03	Dark loam....	.23	2.61 .01	Sandy .....	.34	4.69 .02	Sandy .....	.38	9.47 .04			
10	Dark loam....	.42	13.30 .06	Dark loam....	.33	3.33 .01	Dark loam....	.33	3.33 .01	Sandy .....	.46	10.25 .04			
11	Dark loam....	.49	5.70 .03	Dark loam....	.20	3.00 .01	Dark loam....	.20	3.00 .01	Sandy .....	.32	11.87 .04			
12	Dark loam....	.32	10.00 .03	Dark loam....	.16	2.50 .01	Dark loam....	.16	2.50 .01	Sandy .....	.52	7.88 .04			
Sum of per cents		7.31			8.29			5.60			8.80				
Average per foot		.61	8.56 .05		.69	3.35 .03		.62	4.81 .03		.73	9.20 .06			
Upper 3 feet.*															
Sum of per cents		2.92			4.54			2.20			4.48				
Average per foot		.97	6.90 .06		1.51	3.60 .05		.73	6.10 .05		.49	6.50 .10			

\* Range of most annual plant roots.

There is but little doubt that the humus in both the Cottonwood and Chico soils reaches much deeper than the twelve feet in which the percentage is quite large, for the soil texture is especially favorable for deep development of tree and plant roots, from the decay of which the humus was doubtless derived. In the Chico column there is a sudden increase of humus in the twelfth foot (0.52 per cent), which would indicate former strong development of roots at that point, probably just above a water stratum.

The humus of the Cottonwood column is richest in nitrogen in the upper three feet and shows remarkable and a sudden increased percentage in the tenth foot.

The Chico humus is interesting because of its richness in nitrogen in the lower half of the column.

The Cottonwood and Chico columns alone of the four have humus in sufficient amount and richness to give general averages of more than 0.05 per cent of nitrogen in the entire soil column, and are the only ones having as much as 0.10 per cent of nitrogen in the upper three feet of soil.

*Clay Loams of the Valley.*—These reach south from near Red Bluff and occupy the central part of the Sacramento Valley. As typical of this class of soils, seven columns were obtained from the following localities: three miles west of Tehama, Tehama County; Willows, Glenn County; J. W. Walton's place a few miles south of Yuba City, Sutter County; Woodland, Yolo County; the University Farm, Davis, Yolo County; Live Oak, Sutter County, and from near Elmira, Solano County. In each case a depth of twelve feet was reached, except at Live Oak, where at nine feet the presence of water prevented a deeper sampling.

The clay loams of the Sacramento Valley are generally deficient in humus, as shown by the first foot in each of these seven typical columns and by the results of previous examination of soils from many localities, the average being below 1 per cent. The cause is chiefly continuous grain-growing with shallow cultivation on these lands for thirty or forty years, which has depleted them. This is prominently shown in the soil from the University Farm at Davis which, before purchase by the Univer-

TABLE 5.—HUMUS IN LOAM AND CLAY-LOAM COLUMNS OF THE VALLEY PLAINS

TEHAMA COUNTY				GLENN COUNTY				SUTTER COUNTY				SUTTER COUNTY			
WEST OF TEHAMA				WILLOWS				NEAR LIVE OAK				WALTON'S, SOUTH OF YUBA CITY			
Ft.	Soil Clay 14.60		Humus-Nitrogen in Humus Soil	Soil Clay 21.35		Humus-Nitrogen in Humus Soil		Soil Clay 14.62		Humus-Nitrogen in Humus Soil		Soil Clay 11.67		Humus-Nitrogen in Humus Soil	
	Humus	Humus Soil		Humus	Humus Soil			Humus	Humus Soil			Humus	Humus Soil		
1	Loam	.94	4.47 .04	Clay loam	.98	3.97 .04		Loam	.84	4.90 .04		Loam	.84	4.90 .04	
2	Loam	.78	4.31 .03	Clay loam	.54	3.50 .02		Loam	.44	6.38 .03		Loam	.44	6.38 .03	
3	Loam	.92	4.57 .04	Clay loam	.46	3.25 .02		Loam	.36	3.90 .02		Loam	.36	3.90 .02	
4	Loam	.89	3.78 .03	Clay loam	.40	2.50 .01		Loam	.25	3.45 .01		Loam	.25	3.45 .01	
5	Loam	.50	1.68 .01	Clay loam	.42	4.05 .02		Loam	.22	3.83 .01		Loam	.22	3.83 .01	
6	Loam	.37	2.27 .01	Clay loam	.38	3.69 .01		Loam	.18	4.68 .01		Loam	.18	4.68 .01	
7	Loam	.18	3.11 .01	Clay loam	.36	3.89 .01		Loam	.16	3.51 .01		Loam	.16	3.51 .01	
8	Loam	.16	5.25 .01	Clay loam	.34	4.13 .01		Loam	.13	3.24 .01		Loam	.13	3.24 .01	
9	Loam	.12	4.67 .01	Clay loam	.34	4.13 .01		Loam	.12	2.34 .01		Loam	.12	2.34 .01	
10	Loam	.11	5.09 .01	Clay loam	.32	4.06 .01		Water				Loam	.11	13.51 .02	
11	Loam	.06	....	Clay loam	.28	3.91 .01						Loam	.12	10.83 .01	
12	Loam	.21	....	Clay loam	.28	3.91 .01						Loam	.11	7.62 .01	
Sum of per cents	5.24			5.10				2.70				5.18			
Average per foot	.44	3.92 .02		.43	3.73 .02			.30	4.03 .02			.43	8.98 .03		

Upper 3 feet.\*

Sum of per cents 2.64

Average per foot .88 4.40 .04

\* Range of most annual plant roots.



sity, had been in grain culture for thirty or more years. The humus in its subsoil is nearly double that of the soil and even in the fourth foot is equal to that of the soil itself. A sample of virgin soil taken near this spot was found to have 1.25 per cent of humus, which was probably the original amount in the field, thus showing a loss of nearly 30 per cent. The nitrogen in the virgin soils was 0.07 per cent, but in the cultivated was but 0.04 per cent, which is a loss of more than 40 per cent of nitrogen.

It will be noted that the percentage of humus in the Davis soil below the upper foot is greater throughout its twelve feet than in any other of the columns. This may be accounted for by the greater development of the root systems each year and their subsequent humification. Investigation made by Mr. Farrer, formerly of the University Farm, showed that the roots of wheat, barley, and the California poppy, coincident with those of orchard trees, reached water at a depth of twelve or thirteen feet. If, as is probable, this had been the case for many years, the amount of root material for humification has been large enough for these results. The humus nitrogen of the surface foot is greatest in the Woodland and Yuba City soils, 0.07 per cent and 0.06 per cent respectively; but those columns were taken from an uncultivated lot near Woodland and from the Walton orchard south of Yuba City.

Previous analyses of other clay-loam soils gave the following percentages of humus in the surface foot: Dixon, 1.71; Kell's place near Yuba City, 1.28, and north of Willows, 3.61.

The humus in these columns from the seven localities is not especially rich in nitrogen with the exception of the lower portion of that from Walton's place south of Yuba City; but the amount of humus is so small that the nitrogen given to the soil is very little in amount. The soils from Walton's, Woodland, and Davis are the only ones of the group whose upper three feet have the normal of 0.05 per cent of humus-nitrogen, that of Davis being 0.08 per cent, or approximately 9000 pounds per acre.

*Black Adobe Clay Soils.*—There are several regions of these black clays within the eastern, western, and southern parts of the Sacramento Valley. One of these reaches from southwest

TABLE 6.—HUMUS IN BLACK CLAY ADOBE SOIL COLUMNS, SACRAMENTO VALLEY

	GLENN COUNTY		BUTTE COUNTY		SUTTER COUNTY		SOLANO COUNTY	
	EAST OF WILLOWS		NEAR BIGGS		SOUTHWEST OF YUBA CITY		SOUTH OF DIXON	
	Soil Clay 47.46	Humus- Nitrogen in Humus Soil	Soil Clay 50.09	Humus- Nitrogen in Humus Soil	Soil Clay 39.16	Humus- Nitrogen in Humus Soil	Soil Clay 61.75	Humus- Nitrogen in Humus Soil
1 Black clay....	1.72	5.06 .09	Black clay....	.66 5.89 .03	Black clay....	1.20 4.68 .06	Black clay....	1.05 6.66 .07
2 Black clay....	1.16	4.36 .05	Black clay....	.49 4.58 .02	Gray clay....	.21 5.35 .01	Black clay....	.87 6.09 .05
3 Black clay....	.94	4.98 .05	Black clay....	.40 4.22 .02	Gray clay....	.18 7.80 .01	Black clay....	.77 5.84 .05
4 Black clay....	.62	5.80 .04	Gray clay....	.17 3.30 .01	Gray clay....	.18 7.02 .01	Gray clay....	.55 4.00 .02
5 Black clay....	.36	6.10 .02	Gray clay....	.06 .....	Gray clay....	.11 6.38 .01	Gray clay....	.38 1.58 .02
6 Grayish clay....	.26	9.60 .03	Gray clay....	.08 .....	Gray clay....	.10 9.82 .01	Yellow clay..	.20 4.50 .01
7 Grayish clay....	.22	10.00 .02	Gray clay....	.09 .....	Gray clay....	.08 5.27 tr.	Yellow clay..	.10 1.00 .01
8 Grayish clay....	.20	7.00 .01	Gray clay....	.06 .....	Gray clay....	.12 3.51 tr.	Yellow clay..	.10 .....
9 Grayish clay....	.14	5.70 .01	Gray clay....	.05 .....	Gray clay....	.12 3.51 tr.	Yellow clay..	.10 .....
10 Grayish clay....	.22	3.63 .01	Gray clay....	.04 .....	Gray clay....	.12 3.51 tr.	Yellow clay..	.10 .....
11 Grayish clay....	.20	3.50 .01	Gray clay....	.06 .....	Gray clay....	.13 2.16 tr.	Yellow clay..	.09 .....
12 Grayish clay....	.16	3.76 .01	Gray clay....	.09 .....	Gray clay....	.12 .....	Yellow clay..	.07 .....
Sum of per cents	6.36			2.25		2.67		4.38
Average per foot	.51	5.79 .03		.19 4.49 .02		.22 5.33 .01		.36 4.24 .03
<i>Upper 8 feet: *</i>								
Sum of per cents	3.80			1.55		1.59		2.69
Average per foot	1.27	4.80 .07		.52 4.90 .02		.53 5.90 .03		.89 6.20 .06

\* Range of most annual plant roots.

of Yuba City northward, passing west of Gridley and beyond Biggs. A column was taken from this belt, a few miles southwest of Yuba City, the surface soil of which contained 39 per cent of clay. Below this upper foot the color changed from black to gray. Another column was taken by Mr. F. E. Johnson from near Biggs. This contained 50 per cent of clay in its upper foot and the black color changed to gray below the third foot.

A region of black clay lies three miles east of Willows, Glenn County, whose surface foot contains 47.46 per cent of clay, and the black color extends through five feet. A column of this was taken to the depth of twelve feet.

A few miles south of Dixon, Solano County, there is a large body of very black adobe clay having 61.75 per cent of clay, and the black color passes through three feet, changing to gray below. This soil contains the highest percentage of clay thus far found in any black adobe in the state.

The black adobe clays, with the exception of that from east of Willows, are a disappointment with respect to their humus content, for we had anticipated finding fully 1.5 per cent in each of the upper three or four feet, or hoped that they would at least come up to the average of the other 109 columns of the collection, whose summation for three feet is 2.81 per cent. It is evident from the results that a black color does not always mean a high percentage of humus, for the densely black adobe of Biggs and that south of Dixon each had less than the much lighter colored soils elsewhere, and less than one-half that of the reddish alluvial soil of Chico Creek. The samples from Biggs and Yuba City are from the same belt of black adobe, though many miles apart, and each shows very small percentages below the first foot. Clearly, these clays would be greatly benefited in texture and richness by a good system of green-manuring.

The humus in the adobe soils from the several localities and even in the respective depths of each column is not uniformly rich in nitrogen, as will be seen by reference to the table. It is sometimes richest at a depth of several feet, and frequently there is a sudden and great diminution in an adjoining level, the cause of which is not apparent.



The humus of the adobe soils contains on an average about 5.5 per cent of nitrogen in the first foot and 4.9 per cent in the entire column. The amount in the top soil is greatest in the Willows adobe (0.09 per cent), and least in the Biggs soil (0.03 per cent), or an average of 0.06 for four soils. This is equivalent to 2400 pounds of organic nitrogen per acre in the surface foot which, under the proper ammonifying and nitrifying conditions, is gradually converted into an available supply for plant use.

The humus of the soil near Dixon is richest in nitrogen both in the surface foot and in the upper three feet, though the amount of humus is not so great as in that from Willows.

The amount of humus must be at least 1 per cent, and its nitrogen content must be fair to give to a soil the 0.05 per cent which is regarded as sufficient for present fertility, and we find that of these four localities the soil from Biggs falls short in the first foot and that from Yuba City in the second and third foot respectively.

The percentage in the soils is naturally less and less downward as the humus diminishes, and there are but traces of nitrogen in the lower half of each column. The natural nitrogen supply for the roots of crops must clearly come from the upper three feet in each locality, that from Willows being equivalent to about 8000 pounds per acre in three feet depth.

*Red Mesa and "bedrock" Lands.*—These form a wide border along the eastern and northern sides of the valley, reaching south into San Joaquin County, and are characterized by usually shallow soils underlaid by either heavy, compact red clays or by cemented beds of gravel and grit, forming a hardpan or bedrock at depths of from two to five feet below the surface. Because of the shallowness of these lands, this region is represented by but three columns; one from the bluff of Oat Creek southwest from Red Bluff, and one-half mile west of the Corning road; another from near Sheridan, Placer County, representing the lands on the eastern side of the valley; and the third from near Acampo in San Joaquin County, taken by Mr. F. E. Johnson.

The red soils from Sheridan and from the mesa southwest from Red Bluff show very low percentages of humus, not only

TABLE 7.—HUMUS IN THE SOIL COLUMNS OF RED MESA LANDS

PLACER COUNTY					TEHAMA COUNTY					SAN JOAQUIN COUNTY				
SHERIDAN					SOUTH OF RED BLUFF					ACAMPO				
Ft.	Soil Clay 15.52	Humus	Humus- Nitrogen in		Soil Clay 13.76	Humus	Humus- Nitrogen in		Soil	Humus	Humus- Nitrogen in		Humus	Humus Soil
			Humus	Soil			Humus	Soil			Humus	Humus		
1	Red loam ....	.40	10.52	.04	Red loam ....	.42	6.66	.03	Red loam ....	.60	7.02	.04		
2	Red loam ....	.88	9.21	.04	Red loam ....	.28	5.17	.02	Red loam ....	.18	7.80	.01		
3	Red clay ....	.13	.....	.....	Red loam ..	.22	2.75	.01	Red loam ....	.10	.....	.....		
4	Red clay ....	.10	.....	.....	Red loam ...	.08	1.25	.....	Red loam ....	.10	.....	.....		
5	Light red clay	.05	.....	.....	Red loam ....	.09	.....	.....	Red loam ....	.10	.....	.....		
6	Light red clay	tr.	.....	.....	Red loam ..	.10	.....	.....	Red loam ....	.07	.....	.....		
7	Light red clay	.....	.....	.....	Red loam ....	.07	..	.....						
8	Light red clay	.....	.....	.....	Red loam ....	.07	.....	.....						
9	Light red clay	.....	.....	.....	Red loam ....	.09	.....	.....						
10	Yellow clay..	.....	.....	.....	Red loam ....	.10	...	.....						
Sum of per cents		1.06				1.52				1.15				
Average per foot		.10	.....	.....		.15	.....	.....		.19	.....	.....		
<i>Upper 3 feet:*</i>														
Sum of per cents		.91				.92				.88				
Average per foot		.30	9.85	.04		.31	4.80	.02		.29	.....	...		

\* Range of most annual plant roots.

in the first foot but in each succeeding level down to the tenth foot. The entire amount in each column is but little more than the average of the first foot for the state at large. The darker clay soil from the region of Acampo has more humus in the surface foot and but very little in the lower depths.

The humus of the Sheridan and Acampo soils is rich in nitrogen, but the quantity of humus itself is so small that the amount given to the soil is very little, and below the second foot was inappreciable.

The other soil is peculiar in having a humus that contains very little nitrogen in its third and fourth foot. As a consequence the soil itself is very poor in nitrogen.

All of these soils need good green-manuring with some nitrogen-rich legumes for several successive years to improve their texture and their productiveness.

*Comparison by Classes.*—The average percentages of humus in composite columns of soils of the same class gives an indication of what to expect in lands of these four classes.

TABLE 8.—AVERAGES OF HUMUS IN SOIL TYPES, SACRAMENTO VALLEY

Composite columns	Black adobe clay 4 columns	Stream alluvial loam 4 columns	Clay loam of plains 7 columns	Red mesa and bedrock 3 columns
Humus in first foot .....	1.16	1.47	.99	.47
Sum of, in upper 3 feet .....	2.41	3.54	2.40	.90
Sum of, in entire column .....	3.88	7.50	4.91	1.25
Nitrogen in humus—				
In first foot .....	5.56	5.51	4.68	8.58
Average in upper 3 feet....	5.45	5.78	5.53	7.32
Average in entire column	4.96	6.46	5.03	6.29
Nitrogen in soil—				
In first foot .....	.06	.06	.05	.04
Average in upper 3 feet....	.04	.07	.05	.02
Average in entire column	.02	.04	.02	.01

The figures speak for themselves, and show that the alluvial lands of the streams are richest in humus throughout and that the clay loams of the plains are next, while the black clay, in spite of its color, contains less humus in the upper three feet and in the entire column than the lighter colored plains soil. As a rule, the alluvial lands do not need green-manuring crops, but the others, and especially the red lands, would be greatly benefited thereby.

*Nitrogen in Humus and Soil.*—There is but little difference in the percentage of nitrogen in the humus of the upper three feet of each group, except that in the red soils the figures are highest; but for the entire column the alluvial lands stand at the head, with an average of 6.46 per cent.

For the soil itself the nitrogen is greatest in the stream alluvial, the average in the upper three feet being 0.07 per cent, or approximately 2800 pounds per acre-foot; this is but little more than the minimum amount (0.05 per cent) that is regarded as essential to fertility. Other groups contain less than the alluvial throughout.

#### SOIL COLUMNS OF THE SAN JOAQUIN VALLEY

San Joaquin Valley, with an area of eleven thousand square miles, possesses seven or more large and distinct soil types or regions, each of which is represented in our soil collection.

Twenty-four localities in eight counties were selected from which to secure columns of soil as nearly typical as possible of each region, and fifteen of the columns were taken to depths of

ten or twelve feet, the others being limited in depth either by hardpan, coarse gravel, or other obstruction which prevented the penetration of the soil auger; sometimes the water-table at depths of less than ten feet produced such a mushy condition in the soil as to prevent its being removed by the auger.

*Gray Sandy Loams and Sandy Soils.*—The greater part of the San Joaquin Valley is covered with a grayish sandy, and sandy loam, soil, usually deep and highly productive under irrigation or adequate rainfall. Some of them are highly charged with alkali salts in small areas, but these salts can be removed by proper means of irrigation and subdrainage. Calcareous and black alkali hardpans are also found occasionally, whose injurious effects can be corrected by proper treatment.

From these lands we selected seven localities in five counties from which to secure representative soil columns; three of these contained large amounts of alkali salts and are given a separate discussion. Each of the other columns was taken to a depth of eleven or twelve feet, from the following localities: two miles north of Exeter; three miles west of Tulare; one mile west of Modesto; and near Livingston; taken by Mr. F. J. Randolph.

The soil from west of Tulare has a little more than 1 per cent in the surface, and 2 per cent in the upper three feet, but the others in the table fall far below that percentage, the Modesto and Livingston soils being especially poor. Humus is found throughout the entire column from three of the localities, and contains fair percentages of nitrogen; but the actual amount in the soil itself is very small, except in the upper three feet of the Tulare column. The Livingston soil is little else than sand and is very poor in both humus and nitrogen.

*Gray Clay-Loam Lands.*—The lands of the west side of the San Joaquin Valley, derived from the rocks of the Coast Range and deposited from streams apparently more sluggish than came from the Sierras on the east, contain more clay and are of the clay-loam type of soil. They are more compact than are the sandy soils, and should, therefore, contain more humus than the latter. Three localities where the columns were obtained are on the west side of the valley, one mile south of Tracy, five miles southwest of Los Baños, and one mile west of Mendota.

TABLE 9.—HUMUS IN COLUMNS OF GRAY SANDY LOAMS AND SANDY SOILS, SAN JOAQUIN VALLEY

Fr.	TULARE COUNTY		TULARE COUNTY		STANISLAUS COUNTY		MERCED COUNTY	
	EXETER		WEST OF TULARE		MODESTO		LIVINGSTON	
	Soil Clay 12.17	Humus- Nitrogen in Humus Soil	Soil Clay 8.83	Humus- Nitrogen in Humus Soil	Soil Clay 8.19	Humus- Nitrogen in Humus Soil	Soil Clay 2.85	Humus- Nitrogen in Humus Soil
1	Sandy loam	.65	Sandy	1.22	Sandy	.37	Sandy	.40
2	Sandy loam	.40	Sandy	.98	Sandy	.27	Sandy	.24
3	Sandy loam	.32	Sandy	.44	Sandy	.19	Sandy	.13
4	Sandy loam	.30	Sandy	.32	Sandy	.18	Sandy	.17
5	Sandy loam	.25	Sandy	.30	Sandy	.17	Sandy	.14
6	Sandy loam	.24	Sandy	.29	Sandy	.16	Hardpan	tr.
7	Sandy loam	.23	Sandy	.17	Sandy	.17	Sandy	tr.
8	Sandy loam	.20	Sandy	.17	Sandy	.10	Hardpan	tr.
9	Sandy loam	.16	Gray loam	.09	Sandy	.08	Sandy	tr.
10	Sandy loam	.14	Gray loam	.08	Sandy	.05	tr.	tr.
11	Sandy loam	.15	Gray loam	.07	Sandy	.06	tr.	tr.
12			Gray loam	.09	Sandy	.07	tr.	tr.
	Sum of per cents	3.04		4.22		1.87		1.08
	Average per foot	.38		.35		.15		tr.
	<i>Upper 3 feet: *</i>							
	Sum of per cents	1.37		2.64		.83		.77
	Average per foot	.46		.88		.28		.26

\* Range of most annual plant roots.

TABLE 10.—HUMUS IN COLUMNS OF GRAY CLAY LOAMS, SAN JOAQUIN VALLEY  
 SAN JOAQUIN COUNTY      MERCED COUNTY      FRESNO COUNTY  
 SOUTH OF TRACY      LOS BAÑOS      NEAR MENDOTA

Ft.	Soil Clay 17.08	Humus-Nitrogen in			Soil Clay 15.02	Humus-Nitrogen in			Soil Clay 24.91	Humus-Nitrogen in		
		Humus	Humus	Soil		Humus	Humus	Soil		Humus	Humus	Soil
1	Clay loam.....	1.35	5.20	.07	Clay loam.....	.68	8.64	.06	Clay loam.....	.55	7.10	.04
2	Clay loam.....	.99	5.39	.05	Clay loam.....	.48	9.33	.05	Clay loam.....	.42	6.19	.03
3	Clay loam.....	.83	4.06	.03	Clay loam.....	.44	3.82	.02	Clay loam.....	.38	7.10	.03
4	Clay loam.....	.59	3.81	.02	Clay loam.....	.38	4.05	.02	Clay loam.....	.22	7.27	.02
5	Clay loam.....	.41	4.11	.02	Clay loam.....	.34	4.94	.02	Clay loam.....	.17	7.65	.01
6	Clay loam.....	.25	3.93	.01	Clay loam.....	.28	3.00	.01	Clay loam.....	.12	6.66	.01
7	Clay loam.....	.16	3.51	.01	Clay loam.....	.30	5.60	.02	Clay loam.....	.22	4.09	.02
8	Clay loam.....	.08	3.51	tr	Clay loam.....	.30	9.34	.03	Clay loam.....	.25	2.40	.01
9	Clay loam.....	.10	2.81	tr	Clay loam.....	.24	4.67	.01	Clay loam.....	.40	2.00	.01
10	Clay loam.....	.10	2.81	tr.	Clay loam.....	.22	5.09	.01	Clay loam.....	.30	10.33	.03
11	Clay loam.....	.10	.....	.....	Clay loam.....	.22	6.37	.01	Clay loam.....	.23	2.60	.01
12	Clay loam.....	.08	.....	.....	Gravel .....	.....	.....	..	Clay loam.....	.19	3.15	.01
Sum of per cents		5.04				3.88				3.45		
Average per foot		.42	3.91	.02		.32	5.90	.02		.29	5.55	.02
<i>Upper 3 feet:*</i>												
Sum of per cents		3.17				1.60				1.35		
Average per foot		1.06	4.88	.05		.53	7.26	.04		.45	6.79	.03

\* Range of most annual plant roots.

The Tracy soil is the only one of the above group which contains a fair amount of humus in the first foot and in the three upper feet. But in its lower depths it is much poorer than either that from Los Baños or Mendota. It is interesting to note the distribution downward in the two latter columns, which in the twelfth foot contain one-third of what is in the first foot. The lower six feet of the Los Baños column contains about one-half as much as the upper six feet, while in the Mendota column the upper and lower half are nearly equal in their percentages. The upper half of each column is much richer in nitrogen than the lower, as is the case with all California soils; but there are only traces in the lower six feet of the Tracy column. Deep rooting of plants is thus more favored in the Los Baños and Mendota soils and, with an increased supply of humus in the surface and abundant water, these soils should prove to be fully as productive as that from Tracy.

TABLE 11.—HUMUS IN COLUMNS OF GRAY ALKALI LANDS, SAN JOAQUIN VALLEY

FRESNO COUNTY				TULARE COUNTY				KERN COUNTY				
CENTRAL COLONY				TULARE EXPERIMENT STATION				MIRAMONTE				
Ft.	Soil Clay 4.41	Humus	Humus- Nitrogen in Humus Soil	Soil Clay 10.42	Humus	Humus- Nitrogen in Humus Soil	Soil Clay 13.21	Humus	Humus- Nitrogen in Humus Soil	Soil Clay 10.22	Humus	Humus- Nitrogen in Humus Soil
1	"White Ash"	.45	7.44 .04	Sandy .....	.32	8.78 .03	Sandy loam	.25	5.05 .01	Dark sandy	.18	3.33 .01
2	Loam .....	.37	6.05 .02	Sandy .....	.39	5.04 .02	Sandy loam	.17	4.13 .01	Dark sandy	.10	1.00 tr.
3	Loam .....	.29	2.41 .01	Sandy .....	.40	4.21 .02	Sandy loam	.10	.....	Dark sandy	.14	.71 tr.
4	Whitish loam	.19	5.26 .01	Sandy .....	.28	5.01 .01	Sandy loam	.09	.....	Dark sandy	.17	.59 tr.
5	Whitish loam	.15	4.67 .01	Sandy .....	.12	7.02 .01	Sandy loam	.09	.....	Dark sandy	.18	1.67 tr.
6	Whitish loam	.07	.....	Sandy .....	.10	8.42 .01	Sandy loam	.06	.....	Dark sandy	.10	.....
7	Whitish loam	...	.....	Sandy .....	.10	7.02 .01	Sandy loam	.09	.....	Dark sandy	.08	.....
8	Whitish loam	...	.....	Sandy .....	.13	4.32 .01	Sandy loam	.05	.....	Dark sandy	.07	.....
9	Whitish loam	...	.....	Sandy .....	.06	.....	Sandy loam	.07	.....	Dark sandy	.03	.....
10	Whitish loam	...	.....	Sandy .....	.05	.....	Sandy loam	.05	.....	Dark sandy	.06	.....
11	Whitish loam	...	.....	Sandy .....	.06	.....	Sandy loam	.05	.....	Dark sandy	.05	.....
12	Whitish loam	...	.....	Sandy .....	...	...	Sandy loam	.06	.....	Dark sandy	.04	.....
Sum of per cents				2.01				1.13			1.20	
Average per foot				.13		6.23 .01		.09	.....		.10	.....
<i>Upper 3 feet: *</i>												
Sum of per cents				1.11				.52			.42	
Average per foot				.37		6.01 .02		.17	.....		.14	1.68 .01
* Range of most annual plant roots.												

\* Range of most annual plant roots.

*Gray Alkali Land.*—The three columns of gray sandy and sandy loam valley soils, and the dark loam of Tulare Lake were found to contain high percentages of the several alkali salts, and, therefore, may be put in a class by themselves for consideration.

The "white ash" soil, so called because of its fine, light ashy and silty nature, was taken from near a vineyard in Central Colony, several miles southwest of Fresno, and is the representative of a large region lying on the north side of Kings River and reaching to within two miles of Fresno and eastward toward the foothills of the Sierras. The soils are rich and have been largely devoted to grape-growing. The water-table was reached at a depth of four feet, the soil assuming a white color and, below the sixth foot, losing all traces of humus.

The Tulare column was obtained from the old experiment station tract in a spot where all vegetation had been killed by the alkali salts.

The Tulare Lake column was taken a number of years ago from the bed of the lake, which had long been dried-out to many feet in depth.

The column from Miramonte, fifteen miles west of Wasco, represents what seems to be a belt of low lands or what was once a slough connecting the Tulare Lake with Buena Vista and Kern lakes; in this belt southward the alkali of the lakes had accumulated to great depths. The alkali consists chiefly of the sulfates and chlorids (glaubers and common salt), and in this column was distributed at the rate of one-half of one per cent per foot, giving a total approximating 233,000 pounds for the twelve feet per acre. No vegetation other than scattering alkali weeds was seen.

Analysis of the upper four feet of each of these columns gives the following percentages of alkali salts calculated also to pounds per acre.

TABLE 12.—ALKALI SALTS IN THE UPPER FOUR FEET OF THE COLUMNS

	Percentage in Soil				Pounds per acre; approximate			
	Sulfates	Car- bonates	Chlorids	Total	Sulfates	Car- bonates	Chlorids	Total
Tulare .....	.03	.04	.02	.09	4,800	6,400	3,200	14,400
Central Colony ...	.25	.09	.01	.35	40,000	14,400	1,600	56,000
Miramonte .....	.55	.02	.09	.66	88,000	3,200	14,400	105,600
Tulare Lake bed	.13	.07	.07	.27	20,800	11,200	11,200	43,200



The Fresno and Tulare soils have been under cultivation for the past few years and the alkali has been kept below the surface; as a consequence, root growth was greater and its decay and humification produced more humus than in the Miramonte and Tulare Lake soils. There was also less of alkali salts.

The percentage of humus in the first foot of each of the columns, except that of the lake, is not very much lower than in other gray soils of the Tulare plains. It is distributed through the entire column, except in the water-soaked lower part of the white-ash lands, and the total amount is greater than in some of the alkali-free columns of the valley.

The results, then, apparently show that neither carbonate, sulfate, or chlorid of soda have any injurious effect on humus itself, but that they do so retard or even kill the growth of surface plants and root systems as to cut off the supply of humus-forming material. The humus in the Tulare Lake bed column is exceptionally low in nitrogen from some cause not now apparent, and in all of the soils the nitrogen percentage falls below the minimum required for fertility.

The conclusion is plain, then, that where a green-manure crop can be grown by keeping the injurious alkali salts below a depth of three or more feet by irrigation its conversion to humus under favorable condition is not interfered with.

*Black Adobe Clay and Loam Lands.*—A large area of black adobe land occupies a region in San Joaquin County extending from two miles north of Calaveras River south to French ('amp slough and from the tules or marshes of San Joaquin River eastward toward the hills, and is timbered with oaks. The character of the soil is shown in a column twelve feet in depth taken two and one-half miles southeast of Stockton.

The region continues southward in a narrow belt along the base of the hills into Kern County, being known in Tulare County as "dry bog" because of its tendency to break up into small fragments when dry. It is here underlaid by a reddish clay loam, from which it is sometimes separated by a whitish calcareous and silicious lime and magnesia bed of varying thickness. Two columns of this black clay adobe were secured from near Porterville; one is from the Williams orchard with a depth

TABLE 13.—HUMUS IN COLUMNS OF BLACK CLAYS AND LOAMS, SAN JOAQUIN VALLEY

SAN JOAQUIN COUNTY SOUTHEAST OF STOCKTON				SAN JOAQUIN COUNTY WEST OF TRACY				TULARE COUNTY SOUTH OF PORTERVILLE				TULARE COUNTY WEST OF TULARE			
Ft.	Soil Clay 56.53	Humus- Nitrogen in		Soil Clay 38.80	Humus- Nitrogen in		Soil Clay 29.94	Humus- Nitrogen in		Soil Clay 12.89	Humus- Nitrogen in		Soil Clay 12.89	Humus- Nitrogen in	
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil
1	Black clay...	1.16	7.00 .08	Black clay...	.82	5.10 .04	Black clay...	1.20	3.04 .04	Black loam...	1.16	6.72 .08	Black loam...	1.16	6.72 .08
2	Black clay...	.76	7.38 .06	Black clay...	.70	5.59 .04	Black clay...	1.00	4.91 .05	Black loam...	.62	9.03 .06	Black loam...	.62	9.03 .06
3	Black clay...	.50	7.84 .04	Black clay...	.30	2.50 .01	Black clay...	.83	5.58 .05	Black loam...	.25	8.80 .02	Black loam...	.25	8.80 .02
4	Gray clay...	.22	14.00 .03	Gray clay...	.12	1.69 .01	Marly clay...	.23	7.33 .02	Black loam...	.16	.....	Black loam...	.16	.....
5	Gray clay...	.22	8.91 .02	Gray clay...	.19	.....	Red loam...	.12	10.53 .01	Sandy.....	.11	.....	Sandy.....	.11	.....
6	Gray clay...	.18	3.11 .01	Gray clay...	.09	.....	Red loam...	.06	14.04 .01	Sandy.....	.09	.....	Sandy.....	.09	.....
7	Gray clay...	.11	.....	Gray clay...	tr.	.....	Red loam...	.....	.....	Sandy.....	.09	.....	Sandy.....	.09	.....
8	Gray clay...	.09	.....	Gray clay...	.....	.....	Red loam...	.....	.....	Sandy.....	tr.	.....	Sandy.....	tr.	.....
9	Gray clay...	.08	.....	Gray clay...	.....	.....	Red loam...	.....	.....	Sandy.....	.....	.....	Sandy.....	.....	.....
10	Gray clay...	.07	.....	Gray clay...	.....	.....	Red sandy...	.....	.....	Coarse sand	.....	.....	Coarse sand	.....	.....
11	Gray clay...	.09	.....	Gray clay...	.....	.....	Red sandy...	.....	.....	Coarse sand	.....	.....	Coarse sand	.....	.....
12	Gray clay...	.11	.....	Gray clay...	.....	.....	Sandy.....	.....	.....	Coarse sand	.....	.....	Coarse sand	.....	.....
Sum of per cents		3.59			2.22			3.44			2.48			2.48	
Average per foot		.29			.19			.29			.21			.21	

Upper 3 feet.\*

Sum of per cents 2.42

Average per foot .81

7.40 .06

\* Range of most annual plant roots.

2.03

.64

8.18 .05

of seven feet; the other which appears in the table of analyses is from the Henderson orchard and twelve feet in depth.

On the west side of the valley another narrow black adobe belt reaches along the base of the hills from west of Tracy into Merced County on the south. It is represented by a column of ten feet taken three miles west of Tracy.

Still another belt of black land, not so clayey as the others, though containing from 12 to 14 per cent of clay, lies three miles west of Tulare; a column was taken from it.

It is a matter of much surprise that these clay soils with their very black color should have so small a percentage of humus, the maximum of which is but 1.2 per cent, found in the soil from Porterville. We would naturally anticipate finding fully 10 per cent, and yet in the Tracy soil there is less than 1 per cent. It is evident that the necessary conditions of moisture and warmth for the humification of the vegetable material are not present in these very close, compact clays. The Porterville soil is subjected to irrigation several times annually, while that from Tracy is not, and this may account for the larger amount of humus in the former. In the Stockton column the humus is found to the full depth of twelve feet, while in the others it was found only in the upper six feet.

The nitrogen content of the Stockton soil is good in the upper foot and fair for the three feet, the humus itself being rich in nitrogen, but in the other columns it is quite low and suggests the need of an additional nitrogen supply, either through green-manuring or artificial fertilization.

*Reddish Clay Lands.*—A prominent and wide region of lands of this character occupies a large part of the eastern side of the valley: it is narrow on the north in San Joaquin, Stanislaus, Merced, and Madera counties, and on the south in Tulare and Kern, but quite wide in Fresno County. It is the southern extension of the belt of "bedrock" lands of Sacramento Valley. It is largely characterized by a rocky hardpan of cemented gravel and grit, with thicknesses of an inch to as much as twelve inches and even more, occurring at depths of from two to six or eight feet below the surface. When it occurs near the surface the hardpan gives rise to a rolling or hogwallow feature. An excel-

TABLE 14.—PERCENTAGES OF HUMUS IN COLUMNS OF RED VALLEY LANDS, SAN JOAQUIN VALLEY

SAN JOAQUIN COUNTY				FRESNO COUNTY				FRESNO COUNTY				TULARE COUNTY			
FARMINGTON				EAST OF FRESNO				KEARNEY PARK				LINDSAY			
Ft.	Soil Clay 26.90	Humus- Nitrogen in		Soil Clay 11.01	Humus- Nitrogen in		Soil Clay 14.62	Humus- Nitrogen in		Soil Clay 8.52	Humus- Nitrogen in				
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil			
1	Brown clay....	2.04	6.61 .14	Red loam .....	.85	4.94 .04	Red loam .....	.66	6.81 .05	Red loam .....	.27	3.74 .01			
2	Brown clay....	1.74	7.10 .12	Red loam .....	.70	5.00 .04	Red loam .....	.30	10.33 .03	Red loam .....	.20	5.00 .01			
3	Brown clay....	1.16	9.68 .11	Red loam .....	.32	5.32 .02	Red loam .....	.22	10.00 .02	Red loam .....	.13	6.15 .01			
4	Brown clay....	.64	5.27 .03	Loam .....	.29	2.76 .01	Red loam .....	.25	11.20 .03	Red loam .....	.11	7.27 .01			
5	Light red clay .57		4.93 .03	Loam .....	.56	.....	Red loam .....	.26	10.80 .03	Reddish loam .11	5.50 .01				
6	Light red clay .51		5.51 .03	Loam .....	.16	.....	Red loam .....	.26	10.80 .03	Reddish loam .12	5.83 .01				
7	Light red clay .43		5.88 .03	Loam .....	.....	.....	Red loam .....	.14	14.30 .02	Reddish loam .07	8.57 .01				
8	Light red clay .40		5.62 .02	Loam .....	.....	.....	Red loam .....	.08	17.50 .01	Reddish loam .06	10.00 .01				
9	Light red clay .27		6.24 .02	Loam .....	.....	.....	Red loam .....	.07	.....	Hardpan .....	.....	.....			
10				Loam .....	.....	.....	Red loam .....	.10	.....						
11							Coarse gravel	.....	.....						
12							Coarse gravel	.....	.....						
Sum of per cents		7.76			2.88			2.34			1.07				
Average per foot		.85	6.30 .06		.28	.....		.23	11.47 .03		.13	6.46 .01			
<i>Upper 3 feet:*</i>															
Sum of per cents		4.94			1.87			1.18			.60				
Average per foot		1.65	7.80 .12		.62	5 10 .03		.39	9.05 .03		.20	4.96 .01			

\* Range of most annual plant roots.

lent soil underlies the hardpan, however, and when the latter is broken up by dynamite good results in tree growth are usually obtained.

Columns of this land were obtained from the Butler vineyard a few miles east of Fresno; from the Kearney Park west of Fresno; from Lindsay in Tulare County; and from the place of H. Mueller two miles southwest of Farmington, San Joaquin County. The latter perhaps more properly belongs to the adobe group, though lighter in color.

These lands differ somewhat in their content of clay, that from Farmington being of a clay nature and the other sandy loams. The soil from Farmington contains good humus percentages in the three upper feet and throughout the entire column of ten feet, but each of the other localities shows a deficiency in the surface foot, and throughout the entire depths. In the soil from east of Fresno a hardpan layer was struck in the sixth foot, but was broken up and the soil below it was obtained; no humus was found below the hardpan.

The high humus of the upper three feet of the Farmington column and its high nitrogen content gives to the soil an excellent nitrogen percentage of 0.12 per cent, or approximately 14,000 pounds of organic nitrogen per acre within the range of most plant roots. Bacterial activity in this soil will do much to promote an abundant nitrogen supply for plants and consequent high fertility. The humus of the Kearney Park soil is rich in nitrogen throughout, but the small amount of humus in each foot from the surface down gives but a small amount to the soil, the average for each foot being 0.03 per cent, or about 1200 pounds per acre. The other soil columns are also low in their nitrogen content.

*Delta Plains of Kings and Kern Rivers.*—There are two tracts of this class of lowlands which are made of fine sediment brought down from the Sierra Nevada, the Coast Range on the west contributing little or nothing to these deltas as its streams mainly discharge their sediments westward to the Pacific.

The Mussel Slough region bordering the Tulare Lake receives its sediment from the Kings, Kaweah, and Tule rivers, and covers a very large area. It is timbered with oaks, and the nature

of its soils are shown in columns twelve feet deep taken respectively from near Corcoran and Armona by Mr. F. E. Johnson.

The Kern River delta farther south, with an area of about 290 square miles, is also timbered with oaks. It is represented in the soil collection by a column taken three miles southwest of Bakersfield. Water was reached in the seventh foot.

TABLE 15.—HUMUS IN COLUMNS OF DELTA LANDS, SAN JOAQUIN VALLEY

KINGS RIVER DELTA						KERN RIVER DELTA					
KINGS COUNTY			KINGS COUNTY			KERN COUNTY					
ARMONA			CORCORAN			SOUTH OF BAKERSFIELD					
Ft.	Soil Clay 6.42	Humus-Nitrogen in		Soil Clay 15.11	Humus-Nitrogen in		Soil Clay 11.28	Humus-Nitrogen in			
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		
1	Sandy .....	.46	6.66 .03	Dark loam...	.51	8.26 .04	Loam . . . . .	1.46	4.23 .06		
2	Sandy .....	.32	7.02 .02	Dark loam....	.24	7.02 .02	Loam .....	.71	6.72 .05		
3	Sandy .....	.22	7.66 .02	Dark loam....	.24	8.19 .02	Loam . . . . .	.32	6.87 .03		
5	Sandy .....	.16	7.02 .01	Gray loam....	.24	9.36 .02	Loam .....	.18	6.11 .01		
4	Sandy .....	.20	7.72 .02	Gray loam....	.11	7.66 .01	Loam .....	.40	5.50 .02		
6	Reddish loam	.18	6.24 .01	Gray loam ...	.05	.....	Loam . . . . .	.30	4.66 .01		
7	Reddish loam	.24	7.02 .02	Gray loam....	.06	.....	Loam .....	.26	4.23 .01		
8	Reddish loam	.18	6.24 .01	Gray loam....	.05	.....					
9	Reddish loam	.20	7.02 .02	Gray loam....	.06	.....					
10	Reddish loam	.16	7.02 .01	Gray loam....	.06	.....					
11	Reddish loam	.16	7.02 .01								
12	Reddish loam	.16	7.02 .01								
Sum of per cents		2.64			1.62			3.63			
Average per foot		.22	6.98 .02		.16	8.10 .02		.52	5.48 .03		
<i>Upper 3 feet.*</i>											
Sum of per cents		1.00			.99			2.49			
Average per foot		.33	7.10 .02		.33	7.82 .03		.83	5.90 .04		

\* Range of most annual plant roots.

It was anticipated at the outset that the delta loams of the Tulare Lake and Bakersfield regions would be rich in humus in at least the upper feet, because of their surface vegetation of grasses and weeds and fallen leaves. But humification of these has not taken place thoroughly, and we find that the amount of humus in the upper three feet is not so large as in the sandy loam soils of the plains. The surface soil of the Kern delta is the richest in humus, which gives to the soil its per cent of

nitrogen, but in the three upper feet of each of the columns both humus and nitrogen are below the minimum for fertility.

*River Alluvial Lands and Tule Marshes.*—The rivers of the San Joaquin Valley are usually bordered by narrow bottom lands, the Merced and Kings rivers being exceptions where they enter the valley plains. A column of eight feet depth was taken by Mr. F. E. Johnson from the land of Kings River near Kings River Post Office, and is the only representative of such lands from this valley. The surface soils of other streams have, however, been examined from time to time and their humus content ascertained.

The tule marshes cover a very large region at the junction of the San Joaquin and Sacramento rivers and are divided up into "islands" by many sloughs. The soils of these islands is shallow and peaty though rich, and water appears at but a few feet below the surface. There are localities, however, where a deep soil may

TABLE 16.—HUMUS IN COLUMNS OF LOWLAND SOILS, SAN JOAQUIN VALLEY

RIVER ALLUVIAL FRESNO COUNTY KINGS RIVER P. O.					TULE MARSH SAN JOAQUIN COUNTY STOCKTON			
Ft.	Soil Clay 4.58	Humus	Humus- Nitrogen in		Soil	Humus	Humus- Nitrogen in	
			Humus	Soil			Humus	Soil
1	Sandy . . . . .	1.29	4.25	.06	Black loam . . . .	14.10	5.85	.83
2	Sandy . . . . .	.77	4.38	.03	Black loam . . . .	19.45	4.82	.94
3	Sandy . . . . .	.44	4.47	.02	Very black loam	16.50	5.20	.85
4	Sandy . . . . .	.33	3.83	.01	Very black loam	13.00	4.41	.57
5	Sandy . . . . .	.31	3.62	.01	Lighter loam . . . .	6.92	4.94	.34
6	Sandy . . . . .	.17	4.97	.01	Lighter loam . . . .	2.96	4.35	.13
7	Sandy . . . . .	.17	4.13	.01	Lighter loam . . . .	2.12	5.80	.12
8	Sandy . . . . .	.19	3.72	.01	Lighter loam . . . .	4.28	5.09	.22
9					Clay . . . . .	1.44	10.14	.15
10					Clay . . . . .	.36	2.33	.01
11					Clay . . . . .	.34	2.06	.01
12					Clay . . . . .	.28	6.07	.01
Sum of per cents		3.67				81.75		
Average per foot		.30	4.16	.02		6.81	5.10	.35
<i>Upper 3 feet *</i>								
Sum of per cents		2.50				50.05		
Average per foot		.83	4.40	.04		16.68	3.30	.88

\* Range of most annual plant roots.

be found, and from one of these a column of twelve feet was obtained for us by Mr. W. W. Mackie, then of the United States Bureau of Soils. It was taken from a few miles northwest of Stockton and is an excellent representative of these lands.

The river alluvial is not especially rich in either humus or organic nitrogen, though one would suppose from its alluvial character that the amount of vegetable material in it would be great.

The tule or swamp lands near Stockton have to a large extent been reclaimed by dykes, by pumping out the water, and by protection from overflow. They contain a very large percentage of decayed vegetable matter consisting of tule roots, etc., and this has been quite largely humified, as shown by the tables, especially in the upper four feet. Below this depth the amount suddenly drops from 13 per cent in the fourth to 6.9 per cent in the fifth foot, and from 1.4 per cent in the ninth to 0.36 per cent in the tenth foot. No other tule soil in the state that has been examined contains so high a percentage of humus and organic nitrogen, probably because of extra favorable conditions of warmth and moisture and excess of vegetable matter in the Stockton column. This large amount of humus produces acid soils, and liming is necessary to render them neutral and productive. The 0.83 per cent of humus nitrogen in the surface soil is very great, being equivalent to an average of 25,000 pounds per acre. There is a still greater percentage in the second foot as well as in the third; but below the latter it falls off rapidly to the minimum of 0.01 per cent in the tenth foot.

#### COMPARISON OF SAN JOAQUIN VALLEY SOILS OF DIFFERENT TYPES

The table below gives in a concise form the relative percentages of humus and nitrogen in the eight soil types of the San Joaquin Valley, and from it we can make comparisons more easily than from a study of the soil columns themselves. The types are placed in the order of highest to lowest composite averages of humus in the first foot, and there is almost the same succession in the combined upper three feet and the entire column respectively.



TABLE 17.—PERCENTAGES OF HUMUS AND NITROGEN ACCORDING TO SOIL TYPES,  
SAN JOAQUIN VALLEY

	Tule marsh 1 col.	Stream alluvial 1 col.	Black clays 4 col.	Red lands 4 col.	Delta lands 8 col.	Valley loams 7 col.	Alkali lands 8 col.	Lake bed 1 col.
<i>Humus—</i>								
Per cent in first foot .....	14.10	1.29	1.09	.95	.81	.75	.34	.18
Sum of, in upper 3 feet	50.05	2.50	2.32	2.15	1.49	1.68	.91	.42
Sum of, in entire column	81.75	3.67	2.93	3.51	2.63	3.13	1.58	1.20
Average per foot in upper 3 feet .....	16.68	.83	.82	.72	.49	.56	.30	.14
Average per foot in entire column .....	6.81	.30	.25	.32	.22	.27	.12	.10
<i>Nitrogen in Humus—</i>								
Average in first foot.....	5.85	4.25	5.46	5.52	6.38	6.56	7.09	3.33
Average in upper 3 feet	5.30	4.40	6.09	6.72	6.93	6.09	5.30	1.68
Average in entire column	5.10	4.16	7.10	7.06	6.85	6.10	5.33	1.46
<i>Nitrogen in Soil—</i>								
Average in first foot.....	.83	.06	.06	.06	.04	.05	.03	.01
Average in upper 3 feet	.88	.04	.04	.05	.03	.04	.02	.01
Average in entire column	.35	.02	.03	.03	.02	.02	.01	.01

There are large differences in the amount of humus in the several groups, as is to be expected from soils of such extremely different characters, the highest percentage being in the Stockton tule marshes and the lowest in the strong alkali lands and in the Tulare Lake bed.

The surface soils of the valley are not rich in humus as a rule, and this is well shown in these tables. Even the black clays and loams, which because of their color would be supposed to contain high percentages, were found to have but little more than 1 per cent, and in some instances less than that. Similarly the alluvial and delta soils of Kings and Kern counties are very low in humus. The tule marshes in the region of Stockton are naturally rich, because of the great amount of vegetable matter such as roots and leaves that have accumulated in them, and we find as much as 14 per cent of humus in the first foot and 19 per cent in the second.

*Humus and Nitrogen in the First Foot.*—The general average of humus in the surface soils of the state is 1.25 per cent, and it thus appears that the soils of the San Joaquin Valley fall much below that. In but eight of the columns (omitting the Stockton tule) is there as much as 1 per cent in the surface foot, the

highest being found in the brownish lands of Farmington, and the lowest of 0.18 per cent in the soil of Tulare Lake bed. The general average of all is 0.80 per cent, which is much below the requirements for good texture and productiveness.

The humus of the surface foot contains for the most part a fair percentage of nitrogen, the general average being 5.98 per cent, but there are several soils in which the humus is very poor, and a high amount of such humus is necessary to give to the soil an amount adequate for fertility. It is thought that a fertile soil should not have less than 0.05 per cent of organic or humus nitrogen in the surface foot, and from the tables it is seen that because of the small amount of humus, many of the soils have much less than 0.05 per cent.

The soil richest in nitrogen in the above group is that from Farmington which, because of the abundance and richness of its humus, has 0.14 per cent of nitrogen, equivalent to more than 5000 pounds per acre-foot. The sandy soils of the sandy and black loam plains west of Tulare and the black clay southeast of Stockton each contains from 3000 to 3500 pounds of humus nitrogen per acre-foot.

The humus of the Stockton tule soil contains a fair amount of nitrogen, and the very high percentage of the former gives to the soil the enormous amount of 0.83 per cent of nitrogen, or approximately 32,000 pounds of humus nitrogen per acre-foot. The second foot is even richer.

*Humus in the Upper Three Feet.*—The range of annual plant roots in California soils is in the upper three feet, and this may be considered as the true soil. This combination of three feet more than doubles the amount of humus and its nitrogen that is to be regarded as directly influencing fertility, though the average per foot is lessened. The distribution through the three feet is of greater advantage than if concentrated in the upper foot, for the roots thus secure their nitrogen in a moister soil and away from the heated surface. The general average summation of humus in this depth of three feet for all of the columns, omitting the Stockton tule, is 1.78, or 0.66 per cent per foot, and this is about one-third less than for the state at large. The humus is however fairly rich in nitrogen (6.22 per cent)

and had the former been more abundant the percentage given to the soil would have been sufficient for needs of crops.

*Humus and Nitrogen in the Entire Column.*—Humus was found to occur to depths of ten or twelve feet in but thirteen of the columns. In the brown land of Farmington and the alluvial soil of Kings River it would clearly have been found at that depth had the columns been taken to twelve feet. On the other hand, in five of the ten-foot or twelve-foot columns humus was not found in the lower four or five feet. The percentages diminished downward from the first foot in all cases, in some instances very sharply, and usually added but little to the combined percentage of the upper three feet. The Farmington column with its 7.76 per cent of humus is the richest group (excepting the Stockton marshes), followed by the Tracy loam and Tulare plains soils. Twelve of the columns have higher total amounts of humus than the average of several hundred soils of the humid region, and doubtless the total amount of organic nitrogen in these soils is also greater than that in the humid.

The humus not only varies in its percentage of nitrogen in each of the twenty-five localities from which the soils were taken, but also in the several depths below the surface in each column. It is poorest in the Tulare Lake bed and richest throughout the column from Kearney Park, where each foot except the first contains more than 10 per cent. It is only occasionally that among other columns is there found a humus having as much as 10 per cent. The general average of all is but 6.22 per cent, a figure too low to benefit the soil greatly except where the humus content is above 1 per cent.

The sandy loams of the plains which comprise the greater part of the San Joaquin Valley and the red lands of the eastern side of the valley are but slightly different in their general averages of humus, the latter, because of the high percentages in the Farmington clay, having slightly more throughout its column. The surface soils are clearly in need of green-manure crops that will supplement that humus already present. The soils are liable to form surface crusts where there is so little humus present, and require special care and treatment to prevent injury.

The humus of both plains and the red lands is fairly rich in nitrogen; but the amount for the soil itself is below the normal of 0.05 per cent minimum, except in the first foot. These lands therefore need not only more humus but a humus that is very rich in nitrogen. A leguminous crop, such as the spring vetch, should alone be used for humification, for by it the land secures a greater amount of green stuff, a far greater amount of nitrogen than if grass, rye, or alfalfa were used, and such nitrogen is derived chiefly from the atmosphere.

#### SOIL COLUMNS OF THE LOWER FOOTHILLS OF SIERRA NEVADA

The lower foothills of the Sierra Nevada rising from the valley plains to an elevation of 2500 feet above sea-level forms an important fruit-bearing region. The soils derived from slate and granite are often shallow on the hillsides but deep in the valleys.

TABLE 18.—HUMUS IN SOIL COLUMNS OF FOOTHILLS

BUTTE COUNTY				PLACER COUNTY				AMADOR COUNTY			
PALERMO				NEWCASTLE				EXPERIMENT STATION, JACKSON			
Ft.	Soil Clay 17.80	Humus- Nitrogen in		Soil Clay 14.17	Humus- Nitrogen in		Soil Clay 16.43	Humus- Nitrogen in			
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		
1	Red clay.....	.96	5.85 .06	Reddish loam	1.85	7.45 .10	Red loam ....	1.07	4.50 .0		
2	Red clay.....	.36	8.05 .01	Reddish loam	1.24	9.72 .12	Red loam ....	.54	5.20 .0		
3	Gravelly clay	.20	8.00 .01	Reddish loam	1.18	6.37 .08	Red loam ....	.35	3.61 .0		
4	Gravelly clay	.22	3.64 .01	Reddish loam	.60	12.50 .08	Red loam ....	.35	3.61 .0		
5	Lighter clay..	.12	.....	Reddish loam	.16	8.76 .01	Red loam ....	.25	3.37 .0		
6	Lighter clay..	.10	.....	Gravelly clay	.18	8.32 .02	Granitic clay	.14	4.01 .0		
7	Lighter clay..	.10	.....	Gravelly clay	.34	7.95 .03	Granitic clay	.10	4.21 .0		
8				Gravelly clay	.26	9.63 .03	Granitic clay	.06	4.75 .0		
9				Gravelly clay	.08	.....					
10				Gravelly clay	.12	.....					
11				Gravelly clay	.07	.....					
12				Gravelly clay	.06	.....					
Sum of per cents		2.06			5.64			2.86			
Average per foot		.29	3.91 .02		.47	8.84 .06		.36	4.16 .02		
<i>Upper 3 feet: *</i>											
Sum of per cents		1.52			3.77			1.96			
Average per foot		.51	3.97 .02		1.26	7.80 .10		.65	4.44 .03		

\* Range of most annual plant roots.

Columns of the red slate soil were taken from Palermo, Butte County, and from the former experiment station tract near Jackson, Amador County; also from a bluff near Newcastle, Placer County, taken by Mr. Paul H. Steude of Newcastle.

The red clay soils of Jackson and Palermo are quite similar in the amount of humus in the first foot, but the former is the richer below that. The clay is quite close and compact, and this has prevented the development of roots to the extent permitted by the looser gravelly granitic soil of Newcastle, in which there is more humus. The general average of humus in the first foot is 1.12 per cent; that of thirty-one soils of the foothills previously examined is 1.05 per cent, although it is found to be higher in the valleys farther up in the mountains, in the regions of Auburn, Grass Valley, Nevada City, and Placerville, than near the Sacramento Valley. In percentage summation the general average of the upper three feet of the columns is .80 per cent.

The humus in each of the Palermo and Jackson soils is very poor in nitrogen, not only in the upper three feet but in the entire column; and the nitrogen of the respective soils is also below the normal. On the other hand, the humus in the soil from Newcastle is far richer in nitrogen, there being as much as 8.8 per cent in the humus of each foot of the twelve-foot column. The soil itself contains 0.10 per cent in the upper three feet, which is much above the normal, and is equivalent to about 4000 pounds for each foot in depth per acre.

#### SOIL COLUMNS OF THE COAST RANGE VALLEYS

The Coast Range of mountains, reaching from the Oregon state line south to the Mexican border, has but few agricultural possibilities except in the many valleys enclosed between the mountain ridges. The country north of Mendocino County is especially rugged and the valleys are few, but southward there are many valleys that present splendid agricultural attractions, and we have endeavored to have the soils of the largest and most important ones represented in the columns of this series. Of course, it must be understood that in each valley there are a number of soil variations and gradations from the hills to the lower valley center, and that the column has been selected to

represent the best and most extensive of these, the object being to ascertain to what depth and in what percentage the humus reaches under favorable conditions.

Fifteen valleys in nine counties north of the Santa Ynez Mountains have their soils represented in the series of columns, and in the accompanying tables are arranged in order of occurrence from north to south.

We may conveniently follow the usual subdivision of the Coast Range counties and arrange the table into the counties *north* of San Francisco Bay, embracing seven soil columns; the *bay region* itself, embracing the country east and west of the bay as far south as San José, represented by eight soil columns; and the counties *south* of the bay as far as Santa Barbara, represented by nine soil columns.

#### NORTH OF THE BAY REGION

The valleys represented in this section of the Coast Range are Russian River, Santa Rosa, Los Guillocos, Sonoma, Napa, and Vaca: there are other important though much smaller ones east and west of these and in the counties further north, from which we were unable to secure columns.

*Russian River Valley.*—The soil columns from this valley were taken from the alluvial lands of the hop fields belonging to Mr. T. Boone Miller, six miles south of Healdsburg, and from the red hills three miles southwest of Healdsburg, Sonoma County.

*Santa Rosa Valley.*—The western part of the valley has a heavy adobe soil, which is not so largely in cultivation as the more loamy land of the eastern and middle part. A column of twelve feet depth was taken from the creek alluvial on the Vrooman orchard east of Santa Rosa.

*Los Guillocos Valley.*—This valley is not very wide nor long, and it opens northward into Santa Rosa Valley. Its soil is a reddish loam and is represented by a column taken a short distance southeast of Kenwood, Sonoma County.

*Sonoma Valley.*—The valley opens southward to San Francisco Bay and is largely covered by marsh lands, but the northern

part is higher and comprises better lands. A black adobe clay seems to be the prevailing soil and a column of this was taken near the village of El Verano.

*Napa Valley.*—The soil is chiefly loamy in nature, interspersed with some adobe belts on either side. A column of the former was obtained near Yountville, Napa County, to a depth of twelve feet.

*Vaca Valley.*—This valley is situated among the foothills on the west side of the Sacramento Valley, into which it opens, and is noted for its early fruits. The soil is chiefly a reddish loam, as shown in the column obtained southeast of Vacaville.

These valleys are characterized by having a high humus percentage in the upper foot and also in the four feet which is the usual range of plant roots. The soil from Yountville, Napa Valley, is the richest of the group, and contains nearly 6 per cent of humus in the upper three feet. The Kenwood and El Verano soils are the next in humus content, each containing more than 2 per cent in the first foot and more than 5 per cent in the upper three feet, the range of most plant roots. A notable feature in four of the columns—Russian River, Santa Rosa, El Verano, and Yountville—is that there is more than 1 per cent in each of the upper four feet; in the Santa Rosa column that percentage is found in six feet and almost in the seventh foot. The distribution of humus downward through the entire column of twelve feet is good, the average for the Russian River alluvial being more than 1 per cent for each foot, while the Santa Rosa and Yountville averages very nearly equal it. This is a splendid record and places these soils among the best in the state.

The humus of the Kenwood soil is richer in nitrogen than that of any other column, the average being 10.61 per cent for the seven feet. That of the Santa Rosa has an average of 5.87 in its twelve feet. The richness of the Kenwood soil is chiefly in its lower fourth, fifth, and sixth feet. The humus of the El Verano adobe is for some reason or other weaker in nitrogen than any other (except in its first foot), the general average being but 2.67 per cent in each foot.

The most important consideration, however, is the amount of humus nitrogen occurring in the soil, and we find it to be highest

TABLE 19.—HUMUS IN SOIL COLUMNS NORTH OF THE BAY REGION

RUSSIAN RIVER VALLEY			HEALDSBURG HILLS			SANTA ROSA VALLEY			LOS GILLOS VALLEY		
SONOMA COUNTY			SONOMA COUNTY			SONOMA COUNTY			SONOMA COUNTY		
SOUTH OF HEALDSBURG			SOUTHWEST OF HEALDSBURG			EAST OF SANTA ROSA			KENWOOD		
Ft.	Humus- Nitrogen in		Soil Clay 15.41	Humus- Nitrogen in		Soil Clay 13.33	Humus- Nitrogen in		Soil Clay 26.23	Humus- Nitrogen in	
	Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil
1 Alluvial.....	1.76	4.47 .08	Red loam ...	1.86	4.24 .08	Loam .....	1.95	4.75 .09	Loam .....	2.25	5.80 .13
2 Alluvial.....	1.64	4.00 .06	Red loam ...	1.00	4.20 .04	Loam .....	1.53	5.32 .08	Loam .....	1.95	5.28 .10
3 Alluvial.....	1.34	3.77 .05	Red loam ...	.72	5.13 .04	Loam .....	1.03	7.09 .07	Loam .....	1.01	6.47 .07
4 Alluvial.....	1.24	3.62 .05	Red loam ...	.53	3.58 .02	Loam .....	1.36	5.58 .08	Loam .....	.63	18.10 .11
5 Alluvial.....	1.14	4.06 .05	Red loam ...	.34	4.11 .01	Loam .....	1.23	5.48 .07	Loam .....	.39	10.25 .04
6 Alluvial.....	.89	3.81 .03	Red loam ...	.26	3.07 .01	Loam .....	1.09	4.64 .05	Loam .....	.22	23.20 .05
7 Alluvial.....	.84	4.01 .03	Red loam ...	.14	2.85 .01	Loam .....	.91	5.25 .05	Loam .....	1.64	5.36 .09
8 Alluvial.....	.80	3.71 .03	Yellow loam..	.07	.....	Loam .....	.64	6.14 .04	Gravel .....	.....	.....
9 Alluvial.....	.77	3.30 .03	Yellow loam..	.07	.....	Loam .....	.43	7.18 .03	.....	.....	.....
10 Alluvial.....	.57	2.96 .02	Yellow loam..	.08	.....	Loam .....	.41	7.19 .03	.....	.....	.....
11 Alluvial.....	.63	3.34 .02	Yellow loam..	tr.	.....	Loam .....	.27	6.24 .02	.....	.....	.....
12 Alluvial.....	.66	3.19 .02	.....	.....	.....	Loam .....	.25	5.62 .02	.....	.....	.....
Sum of per cents 12.28				5.07			11.10			8.10	
Average per foot 1.01	3.70 .04			.46	3.89 .03		.92	5.87 .05		1.16	10.61 .08
<i>Upper 3 feet: *</i>											
Sum of per cents 4.74				3.58			4.51			5.21	
Average per foot 1.58	4.08 .06			1.19	4.50 .05		1.50	5.72 .08		1.74	5.80 .10

\* Range of most annual plant roots.



TABLE 19.—HUMUS IN SOIL COLUMNS NORTH OF THE BAY REGION—(Continued)

SONOMA VALLEY				NAPA VALLEY				VACA VALLEY			
SONOMA COUNTY				NAPA COUNTY				SOLANO COUNTY			
EL VERANO				YOUNTVILLE				VACAVILLE			
Ft.	Soil Clay 29.02	Humus- Nitrogen in		Soil Clay 18.60	Humus- Nitrogen in		Soil Clay 18.02	Humus- Nitrogen in			
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		
1	Black clay ....	2.14	4.91 .11	Loam .....	2.64	4.25 .11	Loam .....	1.97	6.39 .13		
2	Black clay ....	1.63	1.47 .02	Loam .....	2.02	4.46 .09	Loam .....	.99	6.35 .06		
3	Black clay ....	1.35	2.59 .04	Loam .....	1.28	3.78 .04	Loam .....	.71	5.90 .04		
4	Black clay ....	1.29	3.88 .05	Loam .....	1.16	2.94 .03	Loam .....	.82	4.04 .03		
5	Reddish clay .79	2.53 .02	Loam .....	Loam .....	.80	3.13 .03	Loam .....	.83	4.34 .04		
6	Reddish clay .69	4.06 .03	Loam .....	Loam .....	.90	3.12 .03	Loam .....	.66	2.38 .02		
7	Reddish clay .52	1.54 .01	Loam .....	Loam .....	.68	2.94 .02	Loam .....	.45	3.11 .01		
8	Reddish clay .64	1.25 .01	Loam .....	Loam .....	.64	2.66 .02	Loam .....	.23	6.08 .01		
9	Reddish clay .60	1.67 .01	Loam .....	Loam .....	.36	3.61 .01	Loam .....	.48	8.12 .04		
10	Reddish clay .40	2.75 .01	Loam .....	Loam .....	.28	3.57 .01	Loam .....	.59	4.23 .03		
11	Gravel .....	.....	Loam .....	Loam .....	.30	3.68 .01	Loam .....	.29	1.72 .01		
12	.....	.....	Loam .....	Loam .....	.32	3.45 .01	Loam .....	.31	1.61 .01		
Sum of per cents 10.05				11.38				8.33			
Average per foot 1.00		2.67 .03	.95		3.46 .03	.71		4.51 .04			
Upper 3 feet.*											
Sum of per cents 5.12				5.94				3.67			
Average per foot 1.71		3.00 .05	1.98		4.16 .08	1.22		6.21 .08			

\* Range of most annual plant roots.

(0.13 per cent) in the surface soil of Vacaville and Kenwood, with the equivalent of about 5000 pounds per acre-foot; the general average of all surface soils being 0.10 per cent, or 4000 pounds per acre-foot. This is a very good amount. In a depth of three upper feet, comprising the range of chief feeding roots of the plant, we again find the highest percentage to be in the Kenwood soil, with an average of 0.10 per cent for each foot, or about 12,000 pounds of humus-nitrogen per acre in the three feet. The Santa Rosa, Yountville, and Vacaville soils are nearly equal in their amounts of 0.08 per cent. In the entire column of twelve feet, the percentage of humus-nitrogen is greatest in the Santa Rosa alluvial (0.05 per cent), while in the Russian River alluvial and the Vaca Valley soils the percentage is 0.04 per cent. This high amount of humus-nitrogen in these soils becomes gradually available to plants only through the action of bacteria.

The soils of the valleys north of the bay region may then be considered as being well supplied in humus and humus-nitrogen, which is well distributed throughout a depth of ten or twelve feet, thus affording special inducements for deep rooting and deep feeding of plants.

#### THE BAY REGION

*Alameda Plains.*—The bay shore rises gently eastward to the foot of the Contra Costa Hills, a distance of about two miles. On this slope the soil is largely of an adobe clay nature. The city of Berkeley is situated on this slope, the University of California being at the foot of the hills. A column of the clay adobe was taken from the economic garden on the University grounds. Southward from Berkeley and Oakland the slope widens into a plain traversed by streams from the Coast Range bordered by wide bands of a more loamy soil, and upon it are found extensive farms. A column of the loam was taken from the land of Mrs. Sanborn, south of Niles, and another from the Meek place near Hayward.

These are excellent soils, with fair humus and humus-nitrogen. The Berkeley adobe is rich in nitrogen throughout.

TABLE 20.—HUMUS IN SOIL COLUMNS OF ALAMEDA PLAINS, ALAMEDA COUNTY

BERKELEY				HAYWARD				NILES			
Ft.	Soil Clay 81.98	Humus- Nitrogen in		Soil Clay 9.21	Humus- Nitrogen in		Soil Clay 10.76	Humus- Nitrogen in			
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		
1	Black clay ....	2.18	6.67 .14	Loam .....	1.81	4.65 .08	Dark loam ....	1.10	5.36 .06		
2	Black clay ....	2.07	5.97 .12	Loam .....	1.04	5.67 .06	Dark loam ....	1.00	4.20 .06		
3	Black clay ....	1.84	7.83 .14	Loam .....	.74	6.26 .05	Dark loam ....	.78	5.04 .04		
4	Black clay ....	1.90	4.68 .09	Loam .....	.80	4.73 .04	Dark loam ....	.56	5.01 .03		
5	Yellow clay..	.95	7.05 .07	Loam .....	.92	4.58 .04	Dark loam ....	.44	5.74 .03		
6	Yellow clay..	1.06	5.37 .06	Loam .....	.78	6.48 .05	Dark loam ....	.38	5.17 .02		
7	Yellow clay..	.48	16.45 .08	Loam .....	.68	6.18 .04	Dark loam ....	.40	5.27 .02		
8	Yellow clay..	.37	24.10 .09	Loam .....	.57	5.26 .03	Dark loam ....	.62	8.15 .05		
9	Yellow clay..	.36	12.50 .05	Loam .....	.38	7.90 .03	Dark loam ....	.70	4.01 .03		
10	Yellow clay..	.36	14.70 .05	Sandy.....	.32	6.58 .02	Dark loam ....	.68	4.54 .03		
11	Yellow clay..	.45	9.35 .04	Dark loam ....	.41	6.85 .03	Dark loam ....	.56	4.51 .03		
12	Yellow clay..	.49	8.16 .04	Dark clay.....	.80	1.28 .01	Dark loam ...	.46	6.10 .03		
Sum of per cents 12.46					9.25			7.68			
Average per foot 1.04					.77	5.53 .04		.64	5.25 .03		
<i>Upper 3 feet: *</i>											
Sum of per cents 6.04					3.59			2.88			
Average per foot 2.01					1.19	5.60 .06		.96	4.90 .05		

\* Range of most annual plant roots.

*East of Contra Costa Hills.*—Eastward across the Contra Costa hills several narrow valleys connect the large and fertile Livermore Valley with the bay shore on the north, and representative columns of soils were taken from three of these.

*Ignacio Valley.*—Along Walnut Creek there is a narrow belt of black clay-loam soil bordered by land more adobe-like in nature which extends to the low mesa and hills. The higher land on the mesa and bordering it in the valley has a stiff and black adobe clay soil about three feet in depth and underlaid by a whitish material. The column of the former was obtained from the place of Professor F. T. Bioletti, one mile north of Walnut Creek, Contra Costa County.

*San Ramon Valley.*—This valley is a continuation southward of Walnut Creek Valley, but wider and with more extensive black adobe soils, a column of which was obtained in the vicinity of San Ramon post office, Contra Costa County.

*Livermore Valley.*—The soil of the valley is a loam while that of the low hills of the west and south is reddish and gravelly. A column was taken to a depth of ten feet from the plain in the Santa Rita region, Alameda County.

TABLE 21.—HUMUS IN SOIL COLUMNS OF VALLEYS EAST OF CONTRA COSTA HILLS

IGNACIO VALLEY CONTRA COSTA COUNTY WALNUT CREEK				SAN RAMON VALLEY CONTRA COSTA COUNTY SAN RAMON				LIVERMORE VALLEY ALAMEDA COUNTY SANTA RITA			
Ft.	Soil Clay 85.02	Humus- Nitrogen in		Soil Clay 41.00	Humus- Nitrogen in		Soil Clay 10.60	Humus- Nitrogen in			
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		
1	Black clay ....	1.42	5.73 .08	Black clay ....	1.23	5.45 .07	Sandy loam ..	.64	8.78 .06		
2	Black clay ....	1.44	4.96 .07	Black clay ....	1.28	4.38 .06	Sandy loam ..	.83	6.03 .05		
3	Black clay ....	1.16	5.81 .06	Black clay ....	1.08	4.17 .05	Sandy loam ..	.55	5.62 .03		
4	Black clay ....	1.12	5.01 .06	Black clay ....	.84	4.29 .04	Sandy loam ..	.45	3.10 .02		
5	Black clay ....	1.08	5.20 .05	Gray clay.....	.81	.....	Reddish loam	.34	3.23 .01		
6	Black clay ....	.70	7.22 .04	Gray clay.....	.75	.....	Reddish loam	.37	3.78 .01		
7	Black clay ....	.60	6.08 .04	Gray clay.....	.29	.....	Reddish loam	.43	2.57 .01		
8	Black clay ....	.60	6.32 .03	Gray clay.....	.20	.....	Sand .....	.24	3.38 .01		
9	Black clay ....	.42	7.85 .03	Gray clay.....	.14	.....	Sand .....	.28	2.14 .01		
10	Black clay ....	.52	6.75 .04	Gray clay.....	.12	.....	Sand .....	.33	4.25 .01		
11	Black clay ....	.36	6.63 .02	Gray clay.....	.09	.....					
12	Black clay ....	.26	5.40 .01	Gray clay.....	.07	.....					
Sum of per cents		9.68			6.90			4.46			
Average per foot		.80	6.04 .04		.57	4.57 .05		.45	4.28 .02		
<i>Upper 3 feet :*</i>											
Sum of per cents		4.02			3.59			2.02			
Average per foot		1.34	5.50 .07		1.19	4.70 .05		.67	6.81 .04		

\* Range of most annual plant roots.

In these two groups of six soils, the adobe clay from Berkeley is the richest in every respect, while that from Walnut Creek is next. High percentages of humus are found to a greater depth in them than in any other of the columns. The Berkeley clay changes color below the fourth foot from black to yellow, while that from Walnut Creek remains dark throughout its twelve feet. The same change from black to gray occurs in the San Ramon adobe below the fourth foot, and each of its upper three feet contains more than 1 per cent of humus. A change from dark to red occurs in the loam soil of Santa Rita in Livermore Valley.

The humus in the Berkeley clay is surprisingly rich in nitrogen—that of the eighth foot reaching 24 per cent—but otherwise the general average of the entire column is 10.20 per cent, which is higher than has been thus far observed in any of the columns except the tule marsh of Stockton. The percentage of nitrogen in the humus of the other five columns is not especially high. For the upper three feet of the Berkeley column the average of humus-nitrogen in the soil is 0.13 per cent, or approximately 5200 pounds for each foot. This is a high amount and is probably largely due to the excellent cultivation the soil has had for years past.

The percentage of humus in the sandy loam soil taken near Santa Rita in the central part of Livermore Valley is surprisingly low in the upper part of the column, and, as a result, the surface was found dry and crusted over to such an extent that it had to be broken up before the auger could be used. Green-manure crops should be grown and turned under for several successive years in this soil, for by this the texture of the soil would be improved, more nitrogen introduced, and better crops obtained. A comparison of the soils of the two sections shows that those of the bay shore are richer in humus and in nitrogen, both in the surface foot, in the surface three feet, and in the entire column, than those east of the Contra Costa hills.

#### SOUTH OF THE BAY REGION

*Santa Clara Valley.*—This valley, reaching from the Bay of San Francisco southward for seventy miles into San Benito County has a variety of soils. Around the bay, back from the salt marshes, there is a black clay adobe. A column of this was taken from the Morse Seed Farm near Santa Clara.

South of San José the lands are more loamy in character and are represented by a column taken from near Gilroy, by Mr. F. E. Johnson.

The valley west of San José has a soil more sandy in nature and more or less gravelly, on which is located extensive orchards. A column of this soil, seemingly representative of this land, was taken from the El Quito ranch south of Saratoga.

TABLE 22.—HUMUS IN SOIL COLUMNS OF SANTA CLARA VALLEY, SANTA CLARA COUNTY

MORSE SEED-FARM, SANTA CLARA				EL QUITO				GILROY			
Ft.	Soil Clay 58.85	Humus-Nitrogen in		Soil Cl- 10.74	Humus-Nitrogen in		Soil Clay 28.05	Humus-Nitrogen in			
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		
1	Black clay ....	4.43	2.78 .12	Dark loam ....	.75	4.80 .04	Black clay ....	2.76	5.60 .15		
2	Black clay ....	8.66	1.15 .04	Dark loam ....	.66	8.78 .08	Black clay ....	2.12	5.17 .11		
3	Black clay ....	2.80	1.80 .03	Dark loam ....	.96	2.08 .02	Black clay ....	1.80	4.68 .08		
4	Gray clay.....	.61	.....	Dark loam ....	.80	2.25 .02	Black clay ....	1.40	4.81 .07		
5	Gray clay.....	.27	.....	Dark loam ....	.68	1.74 .01	Black clay ....	.96	5.27 .05		
6	Gray clay.....	.20	.....	Dark loam ....	.61	2.13 .01	Black clay ....	.70	7.62 .05		
7	Gray clay.....	.62	.....	Dark loam ....	.73	1.92 .01	Dark clay.....	.68	5.78 .04		
8	Gray clay.....	.23	.....	Dark loam ....	.62	2.26 .01	Dark clay.....	.76	6.28 .05		
9	Gray clay.....	.80	.....	Dark loam ....	.25	.....	Dark clay.....	.60	4.68 .05		
10	Gray clay.....	.13	.....	Dark loam ....	.22	.....	Dark clay.....	.62	4.53 .03		
11				Gravel .....	.....	.....	Dark clay.....	.58	4.36 .03		
12							Dark clay.....	.68	8.72 .03		
Sum of per cents 13.25					6.23			13.66			
Average per foot 1.33					.62	2.62 .02		1.14	5.21 .06		
<i>Upper 3 feet: *</i>											
Sum of per cents 10.89					2.37			6.68			
Average per foot 3.63					.79	3.55 .03		2.23	5.15 .11		

\* Range of most annual plant roots.

The two clay soils from Santa Clara and Gilroy are rich in humus to depths of three and four feet respectively, and in the latter the percentage is quite large through the entire column.

In the Santa Clara column there is a sudden and great fall in percentage below the third foot, where the color also sharply changes from very black to gray. In this soil the clay percentage is very high, producing such a compact and severe texture as to prevent the downward distribution of any mass of plant roots.

The El Quito soil, if a true representative of the orchard lands of the west side of the valley, clearly lacks a sufficient supply of humus in the upper surface foot, though lower down in the column the percentage is very fair. A soil from Cupertino also had a low percentage in its surface foot; the humus, however, in both the El Quito and Santa Clara soil is surprisingly poor in nitrogen. A green-manure crop, rich in nitrogen, is clearly needed to produce conditions for high fertility. The Gilroy column has a fair amount of nitrogen.

*Pajaro Valley.*—The soil of the valley proper is a dark loam, while along the river are adobe clays and clay loams, the latter being largely devoted to sugar-beet culture. The valley loam is alone represented in the soil column series. It was taken from the apple-growing section on the Watsonville side of the valley. Another column of soil was taken from Watsonville Heights, one mile northwest of the town. At a depth of eight feet a mass of decomposed granite was reached. Both columns were taken by Mr. F. E. Johnson.

TABLE 23.—HUMUS IN SOIL COLUMNS FROM NEAR WATSONVILLE,  
SANTA CRUZ COUNTY

PAJARO VALLEY WATSONVILLE				HEIGHTS WATSONVILLE			
Ft.	Soil Clay 9.63	Humus-Nitrogen in		Soil Clay 16.50	Humus-Nitrogen in		
		Humus	Humus Soil		Humus	Humus Soil	
1	Brown loam..	1.38	6.29 .09	Dark loam ..	1.92	4.80 .09	
2	Loam .....	.92	4.87 .05	Dark loam ..	1.46	4.60 .07	
3	Loam .....	.94	4.77 .05	Light clay....	.50	9.00 .05	
4	Loam .....	.74	4.54 .03	Yellow clay..	.21	6.70 .01	
5	Loam .....	.60	4.67 .03	Yellow clay..	.11	6.40 .01	
6	Loam .....	.60	4.67 .03	Yellow clay..	.08	7.50 .01	
7	Loam .....	.92	4.57 .04	Gravelly .....	.09	8.90 .01	
8	Loam .....	.86	3.26 .03	Gravelly .....	.09	4.80 .01	
9	Loam .....	.54	5.19 .03	Hardpan .....	.....	.....	.....
10	Loam .....	.62	3.61 .02				
11	Loam .....	.24	4.67 .01				
12	Loam .....	.46	4.26 .02				
Sum of per cents		8.82			4.46		
Average per foot		.74	4.61 .04		.56	6.60 .03	
<i>Upper 3 feet:*</i>							
Sum of per cents		3.24			3.88		
Average per foot		1.08	5.31 .06		1.29	6.13 .07	

\* Range of most annual plant roots.

The soil of the valley is deeper than that of the Heights, but the percentages of humus in the first foot and in the upper three feet are not as great; neither is its humus so rich in nitrogen. The total amount of humus in the Pajaro Valley column is very good in its distribution downward and this, with the sandy loam nature of the soil, gives encouragement to the deep rooting of

plants in their search for food and moisture. The general average of nitrogen in the soil is good in the upper three feet where it is most needed.

*Salinas Valley.*—The lower or northern part of the valley for about fifty miles is from eight to twelve miles wide, but to the southward the valley is very narrow. Two classes of soil are represented in this series: a column of fifteen feet from the sandy loam lands of the west side of the Salinas River at Fort Romie near Soledad; a column of the black adobe lands around King City, Monterey County, was kindly sent by Mr. R. L. Adams, formerly of the Spreckels Beet Sugar Company.

TABLE 24.—HUMUS IN SOIL COLUMNS OF SALINAS VALLEY,  
MONTEREY COUNTY

FT. ROMIE				KING CITY			
Ft.	Soil Clay 6.83	Humus- Nitrogen in		Soil Clay 32.90	Humus- Nitrogen in		
		Humus	Humus Soil		Humus	Humus Soil	
1	Sandy .....	1.08	6.76 .07	Adobe clay ..	1.15	2.69 .03	
2	Sandy .....	.78	8.46 .06	Adobe clay ..	.94	3.43 .03	
3	Sandy .....	.50	7.30 .04	Adobe clay ..	.61	3.90 .02	
4	Sand .....	.33	12.77 .04	Sandy .....	.25	5.04 .01	
5	Loam .....	.78	5.76 .05	Clay loam.....	.52	5.89 .03	
6	Loam .....	1.02	4.96 .05	Clay loam.....	.41	2.73 .01	
7	Loam .....	.62	4.53 .03	Clay loam.....	.43	3.91 .02	
8	Loam .....	.70	5.43 .04	Clay loam.....	.37	3.41 .01	
9	Loam .....	.77	5.32 .04	Clay loam.....	.33	2.97 .01	
10	Loam .....	.77	5.45 .04	Coarse sand ..	.11	6.86 .01	
11	Loam .....	.44	5.00 .02	Coarse sand ..	.00	.....	....
12	Loam .....	.46	4.79 .02	Coarse sand ..	.00	.....	....
13	Loam .....	.36	6.94 .03				
14	Loam .....	.34	7.35 .03				
15	Loam .....	.41	6.88 .03				
Sum of per cents							
	in 15 feet.....	9.31		in 10 feet.....	5.12		
	Average per foot	.62	6.51 .04		.51	3.98 .02	
<i>Upper 3 feet : *</i>							
	Sum of per cents	2.31			2.70		
	Average per foot	.77	7.50 .06		.90	3.34 .03	

\* Range of most annual plant roots.



These soils are very different in texture, that of Fort Romie being of a sandy nature throughout eighteen feet or more, while that from King City is a stiff clay for a depth of three feet, changing to sand and then a clay loam. The effect of the presence of the sand is shown in the sudden diminution of humus in the fourth foot, and the same change is noted in the tenth foot.

The deep rooting of plants is more marked in the Fort Romie soil by the higher percentages of humus in the lower half of the column. The humus and the soil are each also richer in nitrogen than that of the King City clay, which clearly would be greatly benefited in texture and richness by systematic green-manuring with legumes.

The column of soil from Fort Romie was continued to water at fifteen feet and humus found in fair amount in the last foot.

*Arroyo Grande Valley.*—The soil of this narrow but important valley is of a dark and heavy clay loam nature to the depth of

TABLE 25.—HUMUS IN SOIL COLUMNS OF ARROYO GRANDE VALLEY,  
SAN LUIS OBISPO COUNTY

ARROYO GRANDE					ROUTZAHN SEED FARM				
Ft.	Soil Clay 17.99	Humus-Nitrogen in			Soil Clay 28 80	Humus-Nitrogen in			
		Humus	Humus	Soil		Humus	Humus	Soil	
1	Black loam...	2.50	6.46	.16	Black clay ....	3 78	4 31	.16	
2	Black loam....	2.15	4 67	.10	Dark loam ...	1.50	4.96	.07	
3	Black loam....	1.83	5.37	.10	Dark loam ....	1.18	5.95	.07	
4	Black loam....	1.54	5 65	.09	Dark loam ....	1.52	3.88	.06	
5	Dark loam ....	1.64	5.31	.09	Dark loam ....	1.34	4.82	.07	
6	Dark loam ..	1.56	4.68	.07	Light clay.....	.64	5.27	.03	
7	Dark loam ....	1 36	6.30	.09	Light clay.....	1.04	5.67	.06	
8	Dark loam	.88	8.30	.07	Light clay. ....	70	7 22	.05	
9	Light loam ...	.62	6.36	.04	Light clay.....	.84	6.02	.05	
10	Dark clay.....	.82	5.48	.05	Light clay.....	.96	5 56	.05	
11	Dark clay.....	.88	5.11	.05	Light clay.....	.76	5.91	.05	
12	Dark clay.....	.86	5.22	.05	Dark clay.....	1.20	5.75	.07	
Sum of per cents		16 64				15.46			
Average per foot		1.39	5.74	.08		1.29	5.44	.07	
<i>Upper 3 feet:*</i>									
Sum of per cents		6.48				6.46			
Average per foot		2.16	5.50	.12		2.15	5.07	.10	

\* Range of most annual plant roots.

twelve feet and more. Two columns were obtained by Mr. F. E. Johnson, one near the town of Arroyo Grande, the other from the farm of the Routzahn Seed Company, a few miles to the westward, the only apparent difference being a darker color in the surface foot of the seed-farm soil.

The soil from the Routzahn seed-farm in the Arroyo Grande Valley is certainly remarkably rich in humus for an arid soil; for not only does each foot of the upper five feet and also the seventh contain more than 1 per cent, but the twelfth foot has 1.20 per cent and the tenth nearly 1 per cent. The soil from near the town of Arroyo Grande is even richer than that of the seed-farm, for it contains not only more than 1.50 per cent of humus in each of the upper seven feet, but each of the lower five feet lacks but little of having 1 per cent.

The percentages of humus-nitrogen in these soils are also very high, especially in the upper few feet of each column, that of

TABLE 26.—HUMUS IN SOIL COLUMNS OF SANTA MARIA AND LOMPOC VALLEYS,  
SANTA BARBARA COUNTY

SANTA MARIA VALLEY SANTA BARBARA COUNTY WEST OF SANTA MARIA					LOMPOC VALLEY SANTA BARBARA COUNTY BURPEE SEED-FARM				
Ft.	Soil Clay 10.25	Humus	Humus- Nitrogen in		Soil Clay 38.40	Humus	Humus- Nitrogen in		
			Humus	Soil			Humus	Soil	
1	Dark loam ....	1.44	9.57	.11	Dark clay.....	2.50	5.28	.18	
2	Dark loam ....	1.11	4.81	.05	Dark clay.....	1.56	4.62	.07	
3	Dark loam ....	.84	5.52	.05	Dark clay....	1.51	4.90	.07	
4	Light loam ....	.26	9.72	.03	Dark clay.....	1.93	6.12	.12	
5	Light loam ....	.46	7.76	.04	Dark clay.....	1.07	5.82	.06	
6	Light loam ....	.32	8.97	.03	Dark clay.....	1.18	7.28	.09	
7	Light loam ....	.24	9.36	.02	Dark clay.....	1.33	5.94	.08	
8	Gray loam.....	.21	11.36	.02	Dark clay.....	.80	6.13	.05	
9	Gray loam.....	.21	12.03	.03	Dark clay.....	.43	6.27	.03	
10	Gray loam.....	.23	9.77	.02	Dark clay.....	.21	8.58	.02	
11	Gray loam.....	.09	18.72	.02	Dark clay.....	.24	8.75	.02	
12	Gray loam.....	.16	17.15	.03	Dark clay.....	.27	8.53	.02	
Sum of per cents		5.57				13.08			
Average per foot		.46	10.40	.04		1.09	6.50	.05	
<i>Upper 3 feet:*</i>									
Sum of per cents		3.39				5.57			
Average per foot		1.13	6.67	.07		1.86	4.93	.09	

\* Range of most annual plant roots.

the surface foot being 0.16 per cent, or approximately 6400 pounds per acre. This extreme richness in humus and nitrogen, as well as in potash and phosphoric acid, is responsible for the high productiveness and the national reputation the valley enjoys as a vegetable-seed producing region.

*Santa Maria Valley.*—The soil is chiefly a sandy loam of a brownish color and very deep. A column was taken by Mr. F. E. Johnson near the sugar-beet fields five miles west of the town of Santa Maria.

*Lompoc Valley.*—Three chief classes of soils occupy the valley—adobe near the hills, sandy alluvium near Santa Inez River, and an intermediate type of clay loam between them. The Burpee seed-farm is situated on the clay loam lands, and a column of soil was taken there to a depth of twelve feet, at which point water was reached.

Santa Maria Valley soil has a fair amount of humus in the upper three feet and low percentages through the rest of the column, and this humus has high percentages of nitrogen. The smaller amounts in the second and third foot may be due to withdrawal by plant roots. The general average of organic nitrogen in the upper three feet of soil is good, but for the entire column the percentage is low, because of the low amount of humus.

In strong contrast to the sandy loam soil of Santa Maria Valley is the heavier clay of the Lompoc Valley through which flows Santa Inez River. The upper seven feet of the Burpee soil each contains more than 1 per cent of humus, and this, though not especially rich in nitrogen, gives to the soil itself fairly high percentages, the average in the upper three feet being 0.09 per cent, or about 10,000 pounds per acre.

#### SUMMARY OF RESULTS IN COAST RANGE REGION

The soils of the valleys of the Coast Range are remarkably high in their humus content when compared with the other soils of the arid region. The general average of the state is 1.25 per cent in the surface soil, while that for these coast valleys is 1.94 per cent. But especially does this difference appear when we note the many soils in which more than 1 per cent of humus is found

columns there is a decrease downward, indicating that the humification was of plant roots rather than of vegetable matter deposited from overflow as the land was being built up.

A comparison of the results of the examination of the first foot gives an average of 2.08 per cent of humus north of the bay, 1.82 per cent for the bay region, and 1.93 for the region south, while for the three upper feet the averages are 1.55, 1.59, and 1.45 per cent respectively. This shows quite a close agreement in the soils of the valleys of the three divisions.

A glance over the tables brings out the fact that the Santa Clara soil has a higher amount of humus (4.43 per cent) in the first foot than any soil of the state, except the marsh soils, and that nine of the twenty-five columns of the Coast Range have more than 2 per cent in the surface foot, while in all others but two there is more than 1 per cent.

The portion of the soil column lying below the first foot is richer in humus than that of other agricultural regions. There is more than 1 per cent in each of the four upper feet of ten of the twenty-four columns of the Coast Range; in the columns from Santa Rosa, Russian River, Walnut Creek, and Berkeley this percentage extends through five feet, while in the soil from Lompoc and Arroyo Grande it reaches through seven feet.

The humus of the Santa Maria soil is richest in nitrogen, 9.57 per cent, that of Livermore Valley being next with 8.78. The surface soils whose humus is poorest in nitrogen according to the analyses are King City and Santa Clara adobes. The general average for the twenty-four columns is 5.36 per cent.

The percentage of nitrogen in the surface soil is highest in the soils from Arroyo Grande, viz., 0.16 per cent, or approximately 6400 pounds per acre. Gilroy has 0.15 per cent, Berkeley 0.14 per cent, Lompoc, Vacaville, and Kenwood 0.13 per cent each, and Santa Clara, El Verano, Santa Maria, and Yountville each has more than 0.10 per cent.

*Humus in the Upper Three Feet.*—The percentage of humus in the upper three feet has a summation of 10.89 for the Santa Clara, a little more than 6 per cent for Gilroy, Berkeley, and the two Arroyo Grande columns, and 5.57 per cent for Lompoc, but all others are below this.

The humus of the upper three feet of the Fort Romie soil is richest in nitrogen, 7.50 per cent, while that of the Santa Clara adobe has only an average of 1.74 per cent. The general average of all of this portion of the columns is 5.00 per cent, although fourteen of the twenty-four have a higher percentage.

The humus-nitrogen in this portion of the column varies from 0.13 per cent in the Berkeley adobe, 0.11 per cent in the soils from Gilroy and Arroyo Grande, to more than 0.05 per cent in all others except four.

*Humus in the Entire Column.*—Nine of the twenty-four columns have greater amounts of humus in their depths of ten or twelve feet than has been found in any of the columns representing other parts of the state. The two columns from the Arroyo Grande, with sums of 16.74 per cent and 15.56 per cent, have the highest amounts and are followed in the order of highest by Gilroy, Santa Clara, and Lompoc, each with more than 13.00 per cent, and Berkeley, Russian River Valley, Yountville, Santa Rosa, and El Verano, each with more than 11.00 per cent.

The humus of the entire soil columns of the Coast Range valleys is about as rich in organic nitrogen as that of other soils of the state, the highest average percentage being in the Santa Maria soil and the Berkeley adobe, 10.40 and 10.21 per cent respectively; the Kenwood and Lompoc soils are the next in amount, with 7.79 and 6.50 per cent respectively. The Walnut Creek column has an average of 6.04 per cent, but all other averages are below 6 per cent for the entire column. The organic nitrogen of the soil itself varies from 0.08 per cent in the Arroyo Grande, Berkeley, and Kenwood columns, 0.07 per cent in the Arroyo Grande seed-farm to 0.05 per cent in Santa Rosa, Walnut Creek, San Ramon, and Lompoc; but all others have less.

#### SOIL COLUMNS OF SOUTHERN CALIFORNIA

The region known as Southern California embraces that part of the state lying south of the Sierra Madre and Santa Ynez mountains and includes a number of large and fertile valleys and plains.

*Saticoy Plain.*—The long and broad slope in Ventura County, reaching from the mountains southward to the sea-shore and noted for its lima bean culture, is represented by two soil columns, one taken by Mr. J. B. Neff from near Mound School-house, a few miles east of Ventura, and the other from the orange grove of Mr. N. B. Blanchard, at Santa Paula.

TABLE 27.—HUMUS IN SOIL COLUMNS OF SATICOY PLAINS, VENTURA COUNTY

MOUND SCHOOLHOUSE				SANTA PAULA			
Ft.	Soil Clay 14.18	Humus- Nitrogen in		Soil Clay 15.02	Humus- Nitrogen in		
		Humus	Humus Soil		Humus	Humus Soil	
1	Loam.....	1.23	5.14 .06	Dark loam....	1.23	5.61 .07	
2	Loam.....	1.86	4.96 .07	Dark loam....	.84	6.90 .06	
3	Loam.....	.52	5.67 .03	Dark loam....	.53	6.60 .04	
4	Loam.....	.57	6.84 .04	Light loam....	.45	6.00 .03	
5	Loam.....	.54	5.46 .03	Light loam....	.23	11.30 .03	
6	Loam.....	.48	5.27 .03	Light loam....	.21	20.47 .04	
7	Loam.....	.54	4.68 .03	Dark loam....	.56	9.81 .06	
8	Loam.....	.45	4.99 .02	Dark loam....	.59	1.69 .01	
9	Loam.....	.36	4.68 .02	Dark loam....	.52	10.00 .05	
10	Loam.....	.51	4.13 .02	Dark loam....	.38	9.74 .04	
11	Loam.....	.60	3.98 .02	Dark loam....	.33	7.27 .02	
12	Loam.....	.52	4.32 .02	Fine gravel ..	.24	....	
Sum of per cents		7.68			6.11		
Average per foot		.64	5.01 .03		.51	8.67 .04	
<i>Upper 3 feet:*</i>							
Sum of per cents		3.11			2.60		
Average per foot		1.04	5.26 .05		.87	6.37 .06	

\* Range of most annual plant roots.

The soil from Mound is the better of these two from Saticoy Plain, because of the higher percentage of humus in the column below the first foot; its second foot is richer than the first, and the entire amount is greater than in the Santa Paula. But its humus is poorer in nitrogen (5.01 per cent) than that of Santa Paula (8.67 per cent), and the soil itself is not so well supplied. The upper three feet of each column has a fair amount of soil nitrogen.

It would naturally be supposed that the growing of beans for so many years and leaving the roots and stubble in the ground

would have greatly enriched the Mound soil with nitrogen from the leguminous crop, but it seems to have produced but little if any advantage over the orange orchard land; in fact, the humus of the latter is far richer in nitrogen, perhaps because of the use of fertilizers.

*Santa Clara River Delta.*—This comprises a broad region of dark alluvial land and contains more or less alkali salts in places. Water is usually found at a depth of eight or ten feet below the surface. The delta is noted for its sugar-beet culture. A column was taken near Springville by Mr. J. B. Neff to a depth of eight feet, at which water was reached.

*Pleasant Valley Hill Slope.*—This column was taken from the sandy slope of the hills north of the Southern Pacific Railroad station of Camarillo.

TABLE 28.—HUMUS IN SOIL COLUMNS OF HILLSIDE AND DELTA OF SANTA CLARA RIVER, VENTURA COUNTY

PLEASANT VALLEY HILLSIDE, CAMARILLO					RIVER DELTA SPRINGVILLE			
Ft.	Soil Clay 7.01	Humus	Humus- Nitrogen in		Soil Clay 7.72	Humus	Humus- Nitrogen in	
			Humus	Soil			Humus	Soil
1	Sandy .....	.84	6.19	.05	Dark loam....	.96	7.31	.07
2	Sandy .....	.75	4.93	.04	Dark loam....	.58	6.29	.04
3	Sandy .....	.37	....	....	Dark loam....	.42	4.68	.02
4	Sandy .....	.33	....	....	Sandy .....	.20	8.44	.02
5	Sandy .....	.33	....	....	Sandy .....	.14	5.14	.01
6	Sandy .....	.20	....	....	Sandy .....	.18	4.69	.01
7	Sandy .....	.18	....	....	Silty.....	.22	5.11	.01
8	Sandy .....	.12	....	....	Sand .....	.14	5.14	.01
9	Sandy .....	.17	....	....				
10	Sandy .....	.13	....	....				
11	Sandy .....	.13	....	....				
12	Sandy .....	.11	....	....				
Sum of per cents		3.66				2.84		
Average per foot		.30	....	....		.34	5.70	.02
<i>Upper 3 feet: *</i>								
Sum of per cents		1.96				1.96		
Average per foot		.65	....	....		.65	6.20	.04

\* Range of most annual plant roots.

The soil from Springville in the delta of Santa Clara River is not so rich in humus as its dark color would lead one to suppose, and the entire column of eight feet is quite light though the land is highly productive. A soil from near Oxnard previously examined was found to contain as much as 1.60 per cent of humus, and it would seem that the great body of land farther out in the valley is richer than that near the hills. An interesting example of the penetration of plant roots and their humification in a sandy soil is seen in the column from the hill-side slope north of Camarillo station in Pleasant Valley. It is interesting to note that this soil in the three upper feet has the same percentage as that of the Springville column, although so different in color and texture.

*San Fernando Valley.*—The valley of San Fernando lies north of Los Angeles and includes about two hundred square miles. Much of the valley on the east is covered by debris of cobblestones, gravel, and sand washed from the cañons on the northeast,

TABLE 29.—HUMUS IN SOIL COLUMNS OF SAN FERNANDO VALLEY,  
LOS ANGELES COUNTY

SAN FERNANDO				MISSION SAN FERNANDO			
Ft.	Soil Clay 11.15	Humus	Humus- Nitrogen in	Soil Clay 8.56	Humus	Humus- Nitrogen in	
			Humus Soil			Humus Soil	
1	Loam.....	1.08	4.68 .05	Loam.....	.72	5.46 .04	
2	Loam.....	.70	4.81 .03	Loam.....	.54	6.24 .03	
3	Loam.....	.70	4.61 .03	Loam.....	.50	5.05 .03	
4	Loam.....	.54	4.16 .02	Loam.....	.44	6.38 .03	
5	Loam.....	.52	3.51 .02	Loam.....	.36	5.46 .02	
6	Loam.....	.46	3.66 .02	Loam.....	.34	5.37 .02	
7	Loam.....	.40	5.62 .02	Loam.....	.34	5.37 .02	
8	Loam.....	.20	4.21 .01	Loam.....	.36	4.68 .02	
9	Loam.....	.24	3.51 .01	Loam.....	.42	4.85 .02	
10	Loam.....	.28	3.01 .01	Loam.....	.34	5.78 .02	
11	Loam.....	.20	4.21 .01	Loam.....	.30	5.62 .02	
12	Loam.....	.22	8.83 .01	Loam.....	.26	5.40 .01	
Sum of per cents		5.54		4.92			
Average per foot		.46	4.23 .02	.41		5.43 .02	
Upper 3 feet:*							
Sum of per cents		2.48		1.76			
Average per foot		.83	4.70 .04	.59		5.58 .03	

\* Range of most annual plant roots.



but the rest of the valley has sandy and sandy loam soils, with some heavier clays on the south.

Two columns were obtained by Mr. F. E. Johnson—one from the sandy lands about one-half mile north of Fernando and representing the "granitic wash land" at the foot of the hills; the other from lowland near the old Mission two miles west of Fernando.

The two soils from the San Fernando Valley are quite different in their percentages of clay as well as in amounts of humus in the first foot, in the upper three feet, and in the entire column, the advantage being with the soil from near San Fernando. The Mission soil is more sandy because of the wash from the hills. While the humus of the Mission column is the richer in nitrogen, the two are equal so far as the soil is concerned and are much below the requirements for fertility.

The soil from San Fernando probably is similar in character to that of the large olive orchard at Sylmar, a few miles north near the foot of the hills.

TABLE 30.—HUMUS IN SOIL COLUMNS OF SAN GABRIEL VALLEY,  
LOS ANGELES COUNTY

MONROVIA					COVINA				
Ft.	Soil Clay 7.61	Humus	Humus- Nitrogen in		Soil Clay 6.84	Humus	Humus- Nitrogen in		
			Humus	Soil			Humus	Soil	
1	Sandy loam .	.94	3.82	.04	Sandy loam..	.58	5.31	.03	
2	Sandy loam .	.58	3.77	.02	Sandy loam..	.48	4.08	.02	
3	Sandy loam..	.56	3.75	.02	Sandy loam..	.46	4.26	.02	
4	Sandy loam..	.62	3.54	.02	Sandy loam..	.32	5.25	.02	
5	Sandy loam..	.64	3.59	.02	Sandy loam..	.30	5.60	.02	
6	Sandy loam..	.48	3.13	.02	Sandy loam..	.26	5.38	.01	
7	Sandy loam .	.34	3.24	.01	Sandy loam..	.24	5.83	.01	
8	Sandy loam..	.26	3.07	.01	Sandy .....	.18	4.51	.01	
9	Sandy loam..	.28	2.86	.01	Sandy .....	.16	4.38	.01	
10	Sandy loam..	.30	2.67	.01	Sandy .....	.18	3.89	.01	
11	Sandy loam..	.28	2.50	.01	Gravelly .....	.18	3.89	.01	
12	Sandy loam..	.28	2.86	.01	Coarse gravel	....	.....	....	
Sum of per cents		5.56				3.34			
Average per foot		.46	3.24	.02		.28	4.75	.02	
<i>Upper 3 feet:*</i>									
Sum of per cents		2.08				1.51			
Average per foot		.69	3.78	.03		.50	4.55	.02	

\* Range of most annual plant roots.

*San Gabriel Valley.*—This valley lies east of Los Angeles and reaches from the mountains southward to the alluvial plains. It is watered by the San Gabriel River and bordered on the east by the Puente Hills, and the San Antonio debris cone. Its soil is a sandy loam. A column of soil was taken by Mr. F. E. Johnson from the neighborhood of Covina and "represents an average of all of the different soil types." Another column was taken by Mr. Johnson from near Monrovia and represents the heavier type of soil, the greater part of which is sandy and gravelly wash from the hills.

These soils are quite deficient in the necessary amount of humus to keep them in good physical condition and to furnish adequate nitrogen for the plants.

Attention of orchard growers should especially be given to the yearly growing and turning under of the best leguminous crop available—a crop giving large yields of succulent, easily decomposed stems and leaves and containing high percentages of nitrogen. Until a fair supply of humus in the three upper feet are secured by this method, it seems necessary to apply nitrogen fertilizers to the trees with sufficient irrigation water to carry them down to the feeding roots at a depth of several feet below the surface.

*San Bernardino Valley.*—The eastern portion of the valley of Southern California known as the San Bernardino Valley is separated from the western by a range of low hills and by the debris cone of the San Antonio Creek, which issues from its valley just above Pomona and supplies most of the water to Chino Creek which empties into the Santa Ana River. In the past from time to time, a portion of the San Antonio flow has been diverted into the San Gabriel.

The valley covers a large area and comprises a large variety of soil types, a few of which are represented by soil columns. The valley slopes toward the south to the Rincon Basin and Santa Ana River, and is bordered by a rim of mesa land on the north, east, and south.

*The Valley Plains.*—The soils in the central part of the valley are chiefly of a sandy nature, of excellent depth and good fertility. The lowlands are usually highly charged with alkali that

has been brought in by drainage from the higher valley. Several types of soil from the plains are represented in the columns.

*Sandy loam soils* of the central part of the valley are shown in a column taken by Mr. J. W. Mills from the former University of California Experiment Station tract three miles southwest of Ontario.

*Highly sandy soils*, apparently accumulated by wind storms blowing in from the mountain passes on the north, lie in the central part of the valley and are extensively planted in grape vines, the roots of which penetrate downward for very many feet. A column of this type of soil was secured at Guasti from the land of the Italian Vineyard Company.

*Alluvial plains* of the Santa Ana River, known as the Victoria Tract, a few miles east of San Bernardino.

The soils of the valley plain are all quite sandy and contain but small amounts of humus and of nitrogen. The tendency to crust over and to form a sandy hardpan or plowsole near the surface is quite usual with these soils. A succession of good

TABLE 31.—HUMUS IN SOIL COLUMNS OF VALLEY LANDS, SAN BERNARDINO COUNTY

SOUTHWEST OF ONTARIO				VICTORIA TRACT				GUASTI			
Ft.	Soil Clay 7.60	Humus-Nitrogen in		Soil Clay 8.38	Humus-Nitrogen in		Soil Clay 3.81	Humus-Nitrogen in			
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		
1	Sandy .....	.36	4.68 .02	Loam.....	.69	4.92 .03	Sandy .....	.25	3.36 .01		
2	Sandy .....	.22	5.74 .01	Loam.....	1.02	4.62 .05	Sandy .....	.18	4.42 .01		
3	Sandy .....	.19	8.70 .01	Loam.....	.62	2.26 .01	Sandy .....	.14	4.00 .01		
4	Sandy .....	.13	5.40 .01	Loam.....	.37	3.80 .01	Sandy .....	.12	.....	.....	.....
5	Sand .....	.10	4.21 .01	Loam.....	.24	2.92 .01	Sandy .....	.09	.....	.....	.....
6	Sand .....	.08	3.51 .01	Loam.....	.18	2.22 .01	Sandy .....	.08	.....	.....	.....
7	Sand .....	.09	3.12 .01	Sand .....	.16	1.26 .....	Sandy .....	.07	.....	.....	.....
8	Sand .....	.06	.....	Sand .....	tr.	.....	Sandy .....	.11	.....	.....	.....
9	Coarse sand..	.....	.....	Coarse sand..	.....	.....	Sandy .....	.08	.....	.....	.....
10	Coarse sand..	.....	.....	Coarse sand..	.....	.....	Sandy .....	.07	.....	.....	.....
11	Coarse sand..	.....	.....	Coarse sand..	.....	.....	Sandy .....	.07	.....	.....	.....
12	Coarse sand..	.....	.....	Coarse sand..	.....	.....	Sandy .....	.07	.....	.....	.....
Sum of per cents		1.23			3.28			1.33			
Average per foot		.10	4.34 .01		.27	3.00 .01		.11	.....	.....	.....
<i>Upper 3 feet:*</i>											
Sum of per cents		.77			2.33			.57			
Average per foot		.26	4.70 .01		.78	3.93 .03		.19	3.92 .01		

\* Range of most annual plant roots.

green-manure crops grown, turned under deeply, and allowed to humify properly would add much to the value and fertility of these lands.

*Low Lands.*—Near the border of Chino Creek there are low-lying lands forming a wide border of “moist lands” which do not need irrigation. Ten acres of this near Chino formerly formed a part of the Experiment Station. A column from this was taken by Mr. F. E. Johnson. Another column was taken near Pomona from a marsh spot known as a “cienega.” This is a “limited area showing growth of water-loving plants, appearing sporadically in otherwise arid surroundings—usually hill-sides or valley margins—and occasionally giving rise to flowing springs.”—(Rept. Calif. Expt. Sta., 1892–4, p. 185.)

TABLE 32.—HUMUS IN SOIL COLUMNS OF LOWLANDS, SAN BERNARDINO VALLEY

SAN BERNARDINO COUNTY					LOS ANGELES COUNTY				
CHINO					POMONA CIENEGA				
Ft.	Soil Clay 13.44	Humus	Humus- Nitrogen in		Soil Marsh	Humus	Humus- Nitrogen in		
			Humus	Soil			Humus	Soil	
1	Dark silt ....	2.31	4.86	.11	Black loam....	6.26	4.06	.25	
2	Dark silt .....	1.41	4.28	.06	Black loam....	3.12	3.50	.11	
3	Dark silt .....	.84	5.52	.05	Black loam....	1.64	3.12	.05	
4	Dark silt .....	1.05	4.01	.04	Sandy .....	.72	18.30	.13	
5	Blue clay.....	.72	6.14	.04	Sand . . ....	.12	2.33	.01	
6	Blue clay.....	.82	5.65	.05	Sand ... ..	.66	2.12	.01	
7	Blue clay.....	.78	5.38	.04	Coarse sand..	.15	2.00	.01	
8					Blue sand ....	.63	1.59	.01	
9					Blue sand ....	.61	1.31	.01	
10					Blue sand ....	.55	1.82	.01	
11					Blue sand ....	.30	2.32	.01	
12					Blue sand ....	.57	1.41	.01	
Sum of per cents		7.93				15.33			
Average per foot		1.13	5.12	.06		1.28	3.66	.05	
<i>Upper 3 feet *</i>									
Sum of per cents		4.56				11.02			
Average per foot		1.52	4.90	.07		3.67	3.56	.14	

\* Range of most annual plant roots.

In each of these lowland soils the humus content is excellent though not especially rich in nitrogen. The large amount of the former, however, makes up for any low percentage of nitrogen

and gives to the soil an adequate amount, especially in the upper few feet.

The Chino soil was found on previous examination to hold quite high amounts of alkali salts, among which was nitrates of soda and magnesia (Rept. Calif. Expt. Sta., 1894-5, p. 75).

*Mesa Lands.*—The mesa lands that border the valley are largely of a reddish loam type, interspersed on the north side by deposits of sand and gravel from the hillsides.

The red mesa extends eastward up the San Geronio Pass to its summit at Banning, where it forms quite a high plateau above Timoteo Cañon.

A column of the sandy mesa of the north side was taken in the western part of Highlands to a depth of twelve feet, in which the sandy soil continued to the bottom.

A column representing the red clay mesa was taken near Redlands by Mr. F. E. Johnson. The upper five feet was of a reddish sandy loam nature, below which it changes to a sandy clay and finally to a gritty material.

TABLE 33.—HUMUS IN SOIL COLUMNS OF NORTH-SIDE MESA OF  
SAN BERNARDINO VALLEY

HIGHLANDS					REDLANDS				
Ft.	Soil Clay 4.66	Humus	Humus- Nitrogen in		Soil Clay 10.33	Humus	Humus- Nitrogen in		
			Humus	Soil			Humus	Soil	
1	Sandy .....	.72	6.25	.05	Red loam.....	.43	5.88	.03	
2	Sandy . . . .	.46	9.78	.05	Red loam.....	.33	5.09	.02	
3	Sandy .....	.43	7.90	.03	Red loam. ....	.27	5.20	.01	
4	Sandy .. . .	.37	8.10	.03	Red loam.....	.22	5.11	.01	
5	Sandy .. . .	.32	7.18	.02	Red loam.....	.20	5.75	.01	
6	Sandy .....	.28	6.07	.02	Sandy clay ..	.10	8.42	.01	
7	Sandy .. . .	.27	6.29	.02	Sandy clay ..	.10	8.42	.01	
8	Sandy .....	.25	6.80	.02	Sandy clay ..	.10	8.42	.01	
9	Sandy .....	.25	7.60	.02	Gritty .....	.07	7.00	.01	
10	Sandy .....	.22	7.72	.02	Gritty .....	.08	7.00	.01	
11	Sandy .....	.14	10.00	.02	Gritty .....	.06	.....	.....	
12	Sandy .....	.11	17.27	.02	Gritty .....	.07	.....	.....	
Sum of per cents		3.82				2.03			
Average per foot		.32	8.41	.03		.17	6.63	.01	
<i>Upper 3 feet:*</i>									
Sum of per cents		1.61				1.03			
Average per foot		.54	8.00	.04		.34	5.39	.02	

\* Range of most annual plant roots.

The percentage of humus in each of the above soils is very low, especially in that of the red loam of the Redlands mesa. The percentages decrease steadily into the lower portions of each column.

The Highlands sandy soil is the richer of the two, both in humus and nitrogen, and in the entire column except the upper three feet is fairly supplied with each of these.

The Redlands soil, because of its heavier clay nature, would be more benefited by green-manuring than would that of Highlands.

*South-side Mesa.*—On the south side of the valley the mesa rises from near the border of Santa Ana River and extends to the hills at elevations much above the plains. The soil is very generally a reddish clay loam, more or less gravelly, and overlies granitic debris at depths of many feet.

TABLE 34.—HUMUS IN SOIL COLUMNS OF SOUTH-SIDE MESA OF  
SAN BERNARDINO VALLEY

CORONA				RIVERSIDE			
Ft.	Soil Clay 12.87	Humus- Nitrogen in		Soil Clay 11.78	Humus- Nitrogen in		
		Humus	Humus Soil		Humus	Humus Soil	
1	Red loam.....	.71	7.91 .06	Red loam.....	.63	4.00 .03	
2	Red loam.....	.41	6.85 .03	Red loam.....	.30	6.08 .02	
3	Red loam.....	.42	4.68 .02	Red loam.....	.20	6.32 .01	
4	Red loam.....	.25	4.49 .01	Red loam.....	.11	5.11 .01	
5	Gravelly.....	.15	5.62 .01	Red loam.....	.11	5.11 .01	
6	Gravelly.....	.14	6.02 .01	Red loam.....	.12	5.85 .01	
7	Gravelly.....	.17	3.30 .01	Red loam.....	.11	5.11 .01	
8	Gravelly.....	.26	3.24 .01	Red loam.....	.10	4.00 .01	
9	Gravelly.....	.21	4.01 .01	Red loam.....	tr.	.....	
10	Gravelly.....	.17	3.30 .01	Red loam.....	....	.....	
11	Gravelly.....	.18	3.12 .01	Red loam.....	....	.....	
12	Gravelly.....	.30	3.74 .01	Red loam.....	....	.....	
Sum of per cents		3.37		1.68			
Average per foot		.28	4.70 .02	.14		5.19 .01	
Upper 3 feet:*							
Sum of per cents		1.54		1.13			
Average per foot		.51	6.48 .04	.09		5.47 .02	

\* Range of most annual plant roots.

A column from Riverside was taken from Arlington Heights orchard lands on the south, with the assistance of Mr. J. H. Reed, and we were able to reach a depth of eighteen feet with comparative ease.

Another mesa soil column was taken by Mr. F. E. Johnson from nearly one mile south of the town of Corona, and is a good representative of the orchard land of that vicinity. A depth of twelve feet was reached with difficulty because of the presence of much gravel.

The red mesa land on the south side of the valley also contains very low percentages of humus and of nitrogen. This is especially true of the Riverside soil, more than one-half of whose supply is in the upper two feet. Its nitrogen is also very low. The Corona soil is somewhat richer in nitrogen, but in both soils and doubtless on the rest of the mesa, the need of good green-manure crops for the production of humus is very apparent.

*Los Angeles Alluvial Plain.*—The three rivers, Los Angeles, San Gabriel, and Santa Ana, have each brought down from the San Bernardino Mountains large quantities of silt, sand and clay and formed a large body or region of alluvial lands known as the Los Angeles alluvial plains. Each river preserves its own course through these plains and has built up its own alluvial plain with its own material independently of the others. We thus find that the soils of the San Gabriel are of a more sandy nature than those of the Santa Ana, probably because of the more rapid velocity of the river current over a shorter distance; the Santa Ana leaves the mountains east of San Bernardino and flows by a circuitous route along the southern side of the valley and thus seems to have left much of its coarse material behind and deposited chiefly clays and silts in the alluvial plain.

*The San Gabriel Plain* is represented by three columns. One was taken from the place of Dr. S. S. Twombly, south of Fullerton, to a depth of ten feet; another from the place of Mr. J. B. Neff, near Anaheim; while the third column was taken a mile south of Compton.

The land of that plain belonging to the San Gabriel and especially between this and the Santa Ana River is greatly varied in character, as is indicated by these three columns. In some

TABLE 35.—HUMUS IN SOIL COLUMNS OF SAN GABRIEL RIVER ALLUVIAL PLAIN,  
ORANGE COUNTY

FULLERTON				ANAHEIM				COMPTON			
Ft.	Soil Clay 7.94	Humus-Nitrogen in		Soil Clay 5.78	Humus-Nitrogen in		Soil Clay 7.45	Humus-Nitrogen in			
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		
1	Loam.....	.51	10.89 .06	Sandy .....	.88	8.00 .07	Dark loam....	1.50	6.77 .08		
2	Loam.....	.64	7.82 .05	Sandy .....	.83	6.46 .05	Dark loam....	.91	9.16 .05		
3	Loam.....	.25	18.40 .05	Sandy .....	.68	6.91 .05	Dark loam....	.51	6.50 .04		
4	Loam.....	.22	15.45 .03	Sandy .....	.49	8.00 .04	Light loam....	.60	7.47 .03		
5	Loam.....	.22	20.45 .05	Sandy .....	.41	6.09 .03	Dark loam....	.49	5.18 .09		
6	Loam.....	.26	10.77 .03	Sandy .....	.33	6.06 .02	Light loam....	.53	7.25 .04		
7	Loam.....	.33	15.15 .05	Sandy .....	.33	4.58 .02	Light loam....	.64	5.17 .02		
8	Loam.....	.29	19.31 .06	Sandy .....	.25	4.40 .01	Light loam....	.52	7.00 .02		
9	Loam.....	.29	12.41 .04	Sandy .....	.22	.38 .01	Fine sand ....	.38	12.30 .02		
10	Loam.....	.29	19.31 .06	Sandy .....	.24	.28 .01	Fine sand ....	.42	5.80 .02		
11							Dark loam....	.49	4.15 .03		
12							Dark loam....	.32	3.18 .01		
Sum of per cents		3.30			4.66			7.31			
Average per foot		.33	15.00 .05		.46	5.12 .03		.61	6.66 .04		
<i>Upper 3 feet:*</i>											
Sum of per cents		1.40			2.39			2.92			
Average per foot		.47	12.40 .05		.79	7.12 .06		.97	7.50 .06		

\* Range of most annual plant roots.

localities, as around Compton, the soil is of a dark sandy loam nature, quite micaceous and contains more humus than elsewhere. The Compton column is fairly well supplied with humus and nitrogen throughout the depth of twelve feet, and conditions seem to be especially favorable for deep rooting of plants. In other localities there are heavy sandy deposits from old river overflows, the soil of which while quite fertile needs humus to give it a stronger texture.

The Anaheim column was taken from a walnut orchard whose owner had practiced a system of green-manuring for a number of years with good results. While the percentage of humus is still rather low, yet it contains much nitrogen in the upper portion of the column. The sandy loam soils along the border of this alluvial plain near Fullerton, represented by a column a short distance south of town, are surprisingly low in their humus content, but the humus is remarkably rich in nitrogen, not in



isolated levels as often occurs in other soils, but throughout the entire column, the general average being 0.15 per cent, or approximately 6000 pounds of nitrogen per acre-foot through the entire depth. No other column from the entire collection has a similar record. Bacterial activity to render this humus-nitrogen available to plants should make unnecessary the use of nitrogen fertilizers.

*The Santa Ana Alluvial Plain* is represented by two columns taken by Mr. F. E. Johnson. One of these from two miles south of Santa Ana, represents a considerable area of heavy black adobe; the other, from Irvine Station six miles southeast of Santa Ana, represents a lighter clay loam soil. Both of these columns are from the south side of Santa Ana River.

TABLE 36.—HUMUS IN SOIL COLUMNS OF SANTA ANA RIVER ALLUVIAL PLAIN, ORANGE COUNTY

SANTA ANA					IRVINE				
Ft.	Soil Clay 25 01	Humus	Humus- Nitrogen in		Soil Clay 18.31	Humus	Humus- Nitrogen in		
			Humus	Soil			Humus	Soil	
1	Black clay ...	2.42	3.71	.09	Clay loam ....	.70	5.86	.04	
2	Black clay ...	1.44	3.51	.05	Clay loam ....	.50	8.40	.04	
3	Black clay ....	1.02	4.80	.05	Clay loam ....	.44	4.09	.02	
4	Light clay ....	.83	5.90	.05	Clay loam ....	.38	3.42	.01	
5	Light clay ....	.76	2.22	.02	Clay loam ....	.34	1.76	.01	
6	Light clay ....	.46	2.75	.01	Sand .....	.22	1.82	.01	
7	Light clay ....	.31	2.72	.01	Sand ... ..	.19	.63	.01	
8	Light clay ....	.22	2.55	.01	Sand .....	.20	1.50	.01	
9	Sandy .....	.12	2.34	tr.					
10	Sandy .....	.14	3 01	....					
11	Sandy . ....	.14	3.01	....					
12	Sandy .....	.20	4.18	....					
	Sum of per cents	8.06				2.97			
	Average per foot	.67	3.40	.03		.23	3.43	.02	
	<i>Upper 3 feet. *</i>								
	Sum of per cents	4.88				1.64			
	Average per foot	1.63	4.00	.06		.55	6.12	.03	

\* Range of most annual plant roots.

The adobe clay of Santa Ana contains a fair amount of humus, but the humus is very poor in nitrogen throughout the entire column. In spite of the latter fact, however, the per-

centage of nitrogen in the soil is fair, especially in the first foot. The loam soil from Irvine is not so well supplied with humus and nitrogen as could be desired.

### INTERIOR VALLEYS

The immediate coast line is bordered by a wide mesa which extends south to the state line, interrupted by the characteristic deep and narrow valleys or occasional streams. The valley of the San Diego River is wide and affords much land for culture purposes. The mesa reaches eastward in width to the foot of the higher rolling hills, which rise still higher into the San Jacinto Mountains. Among these hills lie numerous valleys, small and large, whose soils are rich and productive.

*Fallbrook Mesa.*—The hills are for the most part somewhat rolling and capable of cultivation whenever the soil is of sufficient depth and extent. Fallbrook affords an example of these cultivable hills and a column of the soil was taken from the hillside vineyard of Loma Ranch south of the town. The red clay loam soil was of varying thickness and underlaid by a mass of disintegrated granite.

*Perris Valley* lies southeast of Riverside at the western foot of the San Jacinto Mountains. It is about ten miles long and six wide, and has a variety of soils, as described by Professor Hilgard in the *Annual Report of the Agricultural Experiment Station* for 1894–95. The heavier soil from the center of the valley, about a mile east of the town of Perris, was selected and a column taken by Mr. F. E. Johnson.

*The Valley of Escondido* is a large and productive valley lying near the mountains. Its soil is a loam. A column was taken to a depth of twelve feet by Mr. F. E. Johnson in the vineyard of C. C. Katzenburger on the north side of the valley.

*El Cajon Valley* lies among the high mountains twenty-nine miles east of San Diego and has an area of six by four miles. The land is a reddish sandy loam and is very productive. A column twelve feet in depth was taken from near the cross-roads by Mr. F. E. Johnson.

TABLE 37.—HUMUS IN SOIL COLUMNS OF INTERIOR VALLEYS

RIVERSIDE COUNTY				SAN DIEGO COUNTY			
PERNIS VALLEY		FALLBROOK HILLS		ESCONDIDO VALLEY		EL CAJON	
Soil Clay 14.59	Humus- Nitrogen in Humus Soil	Soil Clay 4.22	Humus- Nitrogen in Humus Soil	Soil Clay 6.98	Humus- Nitrogen in Humus Soil	Soil Clay 10.45	Humus- Nitrogen in Humus Soil
1 Loam.....	.35 1.66 .05	Red loam.....	.55 11.82 .03	Red loam.....	.47 6.27 .03	Red loam.....	.93 5.92 .05
2 Loam.....	.81 4.16 .03	Red loam.....	.36 3.12 .01	Red loam.....	.30 3.74 .01	Red loam.....	.36 3.88 .01
3 Loam.....	.67 3.69 .02	Red loam.....	.21 4.01 .01	Red loam.....	.12 4.68 .01	Red loam.....	.21 3.82 .01
4 Loam.....	.50 5.62 .03	Red loam.....	.13 2.16 .01	Red loam.....	.09 4.68 .01	Red loam.....	.07 7.00 .01
5 Sandy.....	.19 3.70 .01	Red loam.....	.20 2.81 .01	Sandy loam.....	.08 ..... ..	Red loam.....	.08 7.00 .01
6 Sandy.....	.17 3.80 .01	Red loam.....	.17 2.48 .01	Sandy loam.....	.14 ..... ..	Red loam.....	.07 7.00 .01
7 Sandy.....	.22 3.19 .01	Sandy.....	.16 1.76 .01	Sandy loam.....	.07 ..... ..	Red loam.....	.07 7.00 .01
8 Sandy.....	.13 5.40 .01	Granitic sand debris	.....	Sandy loam.....	.06 ..... ..	Red loam.....	.06 7.00 .01
9 Sandy.....	.14 4.01 .01		.....	Sandy loam.....	.07 ..... ..	Red loam.....	.06 ..... ..
10 Sandy.....	.21 2.67 .01		.....	Sandy loam.....	.06 ..... ..	Red loam.....	.06 ..... ..
11 Sandy.....	.17 3.30 .01		.....	Sandy loam.....	.06 ..... ..	Red loam.....	.06 ..... ..
12 Sandy.....	.10 2.80 .01		.....	Sandy loam.....	.04 ..... ..	Red loam.....	.07 ..... ..
Sum of per cents	3.66		1.78		1.56		2.10
Average per foot	.30 3.62 .02		.15 4.02 .01		.13 4.84 .01		.18 6.08 .01
<i>Upper 3 feet:*</i>							
Sum of per cents	1.83		1.12		.89		1.50
Average per foot	.61 3.17 .03		.37 6.32 .02		.29 4.90 .02		.50 4.54 .02

\* Range of most annual plant roots.

The surface soil of the column taken near Perris is badly deficient in humus, but in each of the three feet below there is a fair percentage. Green-manuring crops are called for. The hill lands of Fallbrook also are low in their percentages of humus and nitrogen and need nitrogen fertilizers (either through nitrates, stable manure, tankage, or dried blood) as well as good green-manure crops.

The column representing the Escondido Valley is very poor in humus and nitrogen, while that of El Cajon is better supplied, though still much below what is needed for good texture and fertility. Crops should respond to nitrate fertilization on both of these soils.

The humus in all of these soils is surprisingly low for valley lands that are as productive as these.

#### SUMMARY FOR SOUTHERN CALIFORNIA SOIL COLUMNS

The following are the averages obtained by combinations of the twenty-six columns, omitting the marsh of the Pomona cienega:

	Per cent
Average humus in surface foot .....	0.88
Average sum of per cents of humus in upper three feet.....	1.98
Average humus in each of upper three feet .....	0.66
Average nitrogen in humus of surface foot .....	5.53
Average nitrogen in humus of upper three feet .....	5.22
Average nitrogen in surface foot of soil .....	0.05
Average nitrogen in upper three feet of soil .....	0.04
Average nitrogen in each of twelve feet of soil .....	0.02

These percentages are all too low and indicate the great need of good green-manuring treatment for a number of years to build up the upper three feet of soil into a high fertility. This is needed more than phosphate fertilization.

There are but six of the twenty-five soils whose surface foot contains more than 1 per cent of humus; there are but five others that have more than 0.75 per cent, and there are five whose humus falls even below 0.50 per cent.

The lands represented by these columns from Southern California are under such continuous cultivation that the surface foot is hardly a proper unit of comparison; a depth of three feet

would be more nearly correct, for in that is usually found the mass of feeding roots, and no disturbing irregular conditions exist. The general average of the total amount in the three upper feet (sum of per cents) of the columns (omitting that of the Pomona cienega) is 1.98, or an average of 0.66 per foot. This is too small.

The Santa Ana adobe and Chino moist land have each above 4.50 and Mound Schoolhouse has 3.11 per cent, but all others fall below the latter. Six of the columns have more than 2 per cent each, while on the other hand three have each a sum of less than 1 per cent in the three feet, or an average of but 0.25 per cent per foot.

The humus in these soils is with a few exceptions not especially rich in nitrogen, and, as a consequence, the soil is but meagerly supplied.

#### SOIL COLUMNS OF THE NORTHEASTERN LAVA-BED REGION

The lava-bed region, comprising a large portion of the northeastern part of the state, reaches north from the Sierra Nevada mountains into Oregon and is a region of lava-bed plateaus and hills interspersed with occasional valleys that are capable of cultivation to a large extent in grain, some fruits, and alfalfa.

*Honey Lake Valley.*<sup>a</sup>—This valley with its large lake lies between the foot of the Sierras and the lava hills on the north, and has an elevation of about 4000 feet above sea-level. It is divided into the Honey Lake Valley proper and the East Side Valley, which extends from the lake eastward into the desert region.

Honey Lake occupies the greater part of the valley and is bordered on all sides by lands which are being brought under cultivation in grain and alfalfa, except on the east, where the soil is of a more sandy and alkali nature. A column of soil was taken to a depth of ten feet from near Standish on the north side of the valley; a large and luxuriant growth of alfalfa covered the adjoining fields.

<sup>a</sup> Described by Professor Hilgard in *Report of Agricultural Experiment Station*, 1891-92, p. 24.

Another column of soil, taken by Professor G. W. Shaw from a strong alkali tract bordering the lake on the east side near Amedee, contained but 0.25 per cent of humus in the first and 0.15 per cent in the second foot but none below that depth.

*East Honey Lake Valley.*—This desert-like region comprises that portion of Honey Lake Valley in Lassen County that reaches eastward from the lake into Nevada at an elevation of about one hundred feet more than the lands around the lake, and lies between hills of lava debris. The width is about fifteen miles, but increases much more toward the state line. The soil of the plain seems to be, for the most part, quite free from alkali salts, which appear only here and there on the surface; but toward the state line at lower levels the alkali is more abundant. On the north side of this plain the soil is quite level and sandy, with a reddish sandy subsoil.

A wide belt of alluvial land borders Skedaddle Creek in a depth of three or four feet near the creek. Beneath this there is, as shown in a well on the place of A. L. Spoon at Stacy Station, two feet of sand and twenty feet of a calcareous clay underlaid in turn by blue sand and clay. A column of soil was taken for examination from this land to a depth of twelve feet.

It is interesting to note here that a well on the place of Mr. Caudle, north of Stacy, exposed three feet of a reddish soil, three feet of sand, seven feet of boulders, and fifteen feet of white calcareous clay; below this appeared blue clay to a depth of 320 feet from the surface, water being reached at that depth.

In the center of the valley the surface of the land is in low ridges, the sandy soil being blown into hillocks, the soil being underlaid by a very compact gray silty soil to ten feet; then beneath that is ten feet of a fine sand, followed by ten feet of a coarse sand in which water is struck in wells. Beneath this lies a blue clay of fifty feet or more in thickness. A column of soil was taken in this land to twelve feet depth, four miles north of Calneva. On the south of the Western Pacific Railroad from Calneva to the mountains the soil is coarsely sandy.

The soil of Stacy is somewhat remarkable in having so high a percentage of humus in its second foot, and especially in having so much throughout the entire column. It compares well with

TABLE 38.—HUMUS IN SOIL COLUMNS OF HONEY LAKE VALLEY, LASSEN COUNTY

EAST HONEY LAKE VALLEY							HONEY LAKE VALLEY				
STACY				NORTH OF CALNEVA			STANDISH				
Ft.	Soil Clay 11.29	Humus- Nitrogen in		Soil Clay 19.50	Humus- Nitrogen in		Soil Clay 14.58	Humus- Nitrogen in			
		Humus	Humus Soil		Humus	Humus Soil		Humus	Humus Soil		
1	Dark loam.....	.64	7.35 .05	Clay loam ....	.25	3.85 .01	Loam.....	.72	8.45 .03		
2	Dark loam.....	1.26	4.68 .06	Clay loam ....	.10	tr. tr.	Loam.....	.33	tr. tr.		
3	Sandy .....	.45	6.22 .03	Clay loam ....	.14	.....	Loam.....	.17	.....		
4	Sandy .....	.47	6.60 .03	Clay loam ....	.17	.....	Whitish loam	.21	.....		
5	Gritty .....	.44	7.05 .03	Clay loam ....	.13	.....	Whitish loam	tr.	.....		
6	Sandy .....	.39	6.40 .03	Clay loam ....	.13	.....	Whitish loam	.....	.....		
7	Loam.....	.34	5.90 .02	Clay.....	.12	.....	Whitish loam	.....	.....		
8	Loam.....	.27	7.40 .02	Clay.....	.10	.....	Whitish loam	.....	.....		
9	Loam.....	.24	8.32 .02	Clay.....	.16	.....	Whitish loam	.....	.....		
10	Loam.....	.24	7.08 .02	Clay.....	.15	.....	Whitish loam	.....	.....		
11	Loam.....	.22	.....	Sand .....	.08	.....	Whitish loam	.....	.....		
12	Loam.....	.20	.....	Sand .....	.07	.....	Whitish loam	.....	.....		
Sum of per cents		5.16			1.60			1.43			
Average per foot		.43	6.70 .03		.13	.....		.13	.....		
Upper 3 feet:*											
Sum of per cents		2.85			.49			1.22			
Average per foot		.78	6.08 .05		.16	.....		.41	.....		

\* Range of most annual plant roots.

many soils of the more favored portions of the state. The humus is fairly rich in nitrogen, but the amount given to the soil is small.

The soil from the region north of Calneva contains very little humus even in the surface foot, but it is found throughout the entire column and is very poor in nitrogen. The humus is apparently derived from the debris and roots of the sagebrush and alkali weeds that grow on the plain.

With an abundant water supply and the turning under and humification of some good legume crop there is no reason why the lands of East Honey Lake Valley at their elevation of 4000 feet above sea-level should not produce crops suitable to that altitude as well as the lands of Imperial Valley which are below sea-level, though climatic conditions naturally would control the kind of crops grown.

The column taken from an alfalfa field near Standish on the north side of the lake is surprisingly low in humus below the

surface foot. The underlying whitish limy loam seems to have restricted the development of plant roots to the upper four feet. The humus is also poor in nitrogen and was probably derived from the meager roots of sage and alkali weeds.

A soil previously taken from the Susanville Meadows had only 0.33 per cent of humus, and another from two miles west of Amedee had but 0.29 per cent.

*Madaline Plains.*—Passing north from Honey Lake Valley across hills covered with beds of lava, we come to the Madaline Plains, which occupy an immense and almost level basin (probably once an inland lake) at an elevation of 5200 feet above sea-level. Its area is approximately 150 square miles, very irregular in outline, and bordered on all sides by lava hills. Its soil is a dark and very compact clay, underlaid at three or more feet by a light-colored marl of a hardpan nature and upwards of seventy-five feet in thickness, as shown in well-borings. A column of this soil four feet in depth was obtained from the plain to the westward of the place of W. C. Brockman. Previous analyses of other samples show fair amounts of phosphoric acid. Grass and grain are said to do well on this plain. The surface foot was found to contain 0.52 per cent of humus, and the second foot 0.60 per cent, but below this the clay was almost free from it. The soil contains about 0.04 per cent of humus-nitrogen.

*Pitt River Valley.*—Pitt River, with its source at the western foot of the Warner range of mountains, passes through a number of valleys as it flows westward into the Sacramento River. The town of Alturas is located in one of these valleys at the junction of the two forks of the river. The soil of the valley is chiefly meadowland with water at a depth of a few feet and partly grown in tules, but affording large alfalfa tracts. A column of six feet was taken from an alfalfa field near the town. The valley is bordered by lava-beds and hills. Goose Lake Valley to northward and reaching into Oregon has a similar meadowland soil reaching from the lake eastward to the foot of the mountains, where the disintegrated debris affords some higher land on which orchards are planted.

*Klamath Lake Marshes.*—A column from the tule marshes of Klamath Lake, Butte Valley, was obtained for us by Mr. L. S. Robinson for examination.



TABLE 39.—HUMUS IN SOIL COLUMNS OF PITT RIVER AND BUTTE VALLEYS

PITT RIVER VALLEY MODOC COUNTY					BUTTE VALLEY SISKIYOU COUNTY				
ALTURAS					KLAMATH				
Ft.	Soil Clay 29.60	Humus	Humus- Nitrogen in		Soil Clay 11.00	Humus	Humus- Nitrogen in		
			Humus	Soil			Humus	Soil	
1	Clay.....	3.05	1.93	.06	Marsh loam..	3.75	9.15	.34	
2	Clay.....	1.45	3.39	.05	Marsh loam..	3.20	7.10	.23	
3	Clay.....	1.01	5.85	.06	Marsh loam..	3.02	9.07	.27	
4	Clay.....	.87	3.22	.03	Marsh loam..	2.84	8.88	.25	
5	Clay.....	.51	1.09	.01	Ashy loam† ..	.91	7.70	.07	
6	Clay.....	.34	1.65	.01	Ashy loam† ..	.74	7.58	.06	
7	Water ..	....	....	....	Dark clay ....	.06	....	....	
8					Dark clay ...	tr.	....	....	
Sum of per cents		7.23				14.52			
Average per foot		1.20	2.85	.03		1.81	8.25	.20	
<i>Upper 3 feet.*</i>									
Sum of per cents		5.51				9.97			
Average per foot		1.84	3.72	.06		3.32	8.44	.28	

\* Range of most annual plant roots.

† Infusorial earth.

Both of the above soils are rich in humus, though differing greatly in other respects. The Alturas is a clay soil under cultivation in alfalfa, and while its humus is poor in nitrogen the amount given to the soil is fair. On the other hand, the Klamath column is from tule-marsh lands, containing a larger proportion of decaying vegetable matter, and consequently is quite rich in humus. The latter is especially rich in nitrogen and as a consequence the soil is also well supplied in nitrogen through its upper four feet. The fifth and sixth foot are largely made up of diatomaceous earth in which the humus has suddenly diminished to less than 1 per cent.

*Surprise Valley.*—Eastward from Alturas, the Warner range of mountains separates the Pitt River Valley from Surprise Valley with its three large lakes. The land of the eastern side of this valley is strongly charged with alkali salts, but on the west the broad slope from the mountains to the lake presents excellent agricultural capabilities, the production of alfalfa seed being quite a prominent industry. A column of soil ten feet deep

(to water) was obtained two miles south of Cedarville with the assistance of Mr. W. L. Turner, and another column of four feet from the meadowland that forms a broad border to the lake, and in which water was struck at four feet.

TABLE 40.—HUMUS IN SOIL COLUMNS OF SURPRISE VALLEY, MODOC COUNTY

CEDARVILLE				CEDARVILLE			
Ft.	Soil Clay 10.97	Humus	Humus-Nitrogen in Humus Soil	Ft.	Soil	Humus	Humus-Nitrogen in Humus Soil
1	Loam.....	4.27	1.14 .05		Meadow loam	2.71	3.46 .09
2	Loam.....	1.25	3.36 .04		Meadow loam	2.10	4.62 .10
3	Loam.....	1.21	2.81 .08		Meadow loam	.86	8.02 .07
4	Loam.....	.98	2.26 .02		Meadow loam	.79	7.59 .06
5	Loam.....	.64	2.66 .02		Water .....	.....	.. .. ...
6	Loam.....	.63	2.67 .02				
7	Loam.....	.56	3.93 .02				
8	Loam.....	.60	2.33 .01				
9	Loam.....	.51	2.55 .01				
10	Loam.....	.36	4.72 .02				
Sum of per cents		10.96				6.46	
Average per foot		1.10	2.84 .03			1.62	5.92 .08
<i>Upper 3 feet:*</i>							
Sum of per cents		6.73				5.67	
Average per foot		2.25	2.44 .04			1.89	5.37 .09

\* Range of most annual plant roots.

The surface soil of the Cedarville loam is surpassed in its percentage of humus only by the black clay of Santa Clara and the marsh lands among all of the soils of the state thus far examined, and it ranks fifth with regard to the amount in the three upper feet. The percentage throughout the entire column of ten feet is quite high. This high humus content may be due to the alfalfa crops that have been produced for seed on this soil for a number of years past, as a soil from another locality near Cedarville examined ten years ago contained but 1.56 per cent in the surface foot. The humus is, however, remarkably weak in nitrogen, if such was its origin.

The meadow soil lying at a lower level is very similar to that of Alturas in its humus content.

## SOIL COLUMNS OF THE "DESERT" PLAINS

The term "desert" is here applied to the extent of country with scant rainfall and having a vegetation of sagebrush and largely devoid of grasses; a country whose soils are usually rich in the mineral elements of plant food and which are remarkable for their fertility when properly cultivated and abundantly irrigated. The reason for this fertility lies partly in the fact that the humus, though small in amount, is with its nitrogen well distributed throughout a depth of twelve feet and more, in a soil whose sandy or silty texture permits deep rooting of plants.

The desert lands are interspersed with mountain ranges separated by valleys of greater or lesser extent. Some of these valleys have with irrigation been brought under cultivation and settlement and from these a few soil columns were secured.

*Imperial Valley.*—This newly settled part of the state, once the bed of Salton basin which on drying-up became a desert until reclaimed by irrigation, largely lies below the level of the sea. Its soils, derived from the sediment of the Colorado River,

TABLE 41.—HUMUS IN SOIL COLUMNS OF IMPERIAL VALLEY,  
IMPERIAL COUNTY

IMPERIAL				EL CENTRO			
Ft.	Soil Clay 50.43	Humus	Humus- Nitrogen in	Soil Clay 47.42	Humus	Humus- Nitrogen in	
			Humus Soil			Humus Soil	
1	Silty clay. . .	.26	6 14 .02	Silty clay.....	.30	5.15	.02
2	Silty clay.....	.24	3.51 .01	Silty clay.....	.23	5.49	.01
3	Silty clay.....	.21	3.34 .01	Clay.....	.22	6.38	.01
4	Clay.....	.19	4.43 .01	Clay.....	.21	4.54	.01
5	Silt .....	.17	6.61 .01	Clay.....	.32	4.39	.01
6	Silt .....	.21	4.01 .01	Clay. ....	.27	5.20	.01
7	Clay.....	.22	3.83 .01	Clay.....	.26	4.86	.01
8	Clay. ....	.15	4 68 .01	Clay.....	.31	4.08	.01
9	Silt .....	.14	4.01 .01	Clay. ....	.28	5.01	.01
10	Silt .....	.13	5.40 .01	Clay.....	.32	3.51	.01
11	Silt .....	.10	8.42 .01	Clay.....	.29	2.91	.01
12	Clay.....	.14	4.01 .01	Clay.....	.20	5.62	.01
Sum of per cents		2.16		3.21			
Average per foot		.18	4.86 .01	.27		4.76	.01
<i>Upper 3 feet:*</i>							
Sum of per cents		.71		.75			
Average per foot		.24	4.38 .01	.25		5.67	.01

\* Range of most annual plant roots.

have a peculiar light pinkish tint and are made up of alternating strata of silt and a very plastic and impervious clay of varying thicknesses. When this clay forms the surface soil, the difficulties of cultivation and irrigation are very great; but if the clay lies at a depth of several feet below the surface of a silty soil then the reverse is true. This peculiar type of land is represented in the series by two columns of soil, one from the vicinity of Imperial and the other from near El Centro; these and that from Coachella were taken by Mr. F. E. Johnson.

*Mojave River Mesa.*—The mesa or plain is formed by accumulation of granitic debris from the Sierra Madre Mountains on the south and reaches far out toward Barstow, San Bernardino County. This debris is coarse and quite compact, and in the neighborhood of Victorville is said to have a thickness of thirty-five feet or more. It is here overlaid by about three feet of a gray sandy soil, also quite compact and supporting a sparse vegetation of weeds. A column of eleven feet was obtained three miles west of Victorville. The soil was found to contain but 0.13 per cent of humus in the surface foot, 0.14 per cent in the second, 0.10 per cent in the third, and 0.08 per cent in the coarse sand of the fourth foot. There were but traces of nitrogen in the humus. The soil has 11.05 per cent of clay.

*Coachella Valley.*—A column was obtained from the vicinity of Coachella and represents the higher and more sandy lands that bordered the old Salton Lake of the Imperial country.

*Owens River Valley.*—The agricultural lands of this valley lie chiefly on the west side of Owens River and are formed of the debris of the adjoining Sierra Nevada. A column of soil was secured through the kindness of Mr. W. K. Winterhalter of San Francisco.

It was to be anticipated that the soils of what are known as the desert plains of the eastern and southeastern part of the state would be very poor in humus because of their very sandy nature, together with small rainfall, extreme summer heat and scant vegetation, but it was a surprise to find that humus occurred at depths of twelve or more feet below the surface. The soil poorest in humus is that from the mesa plain just west of Victorville, in which plant roots were found in a depth of three feet only, the underlying material being too compact for root

TABLE 42.—HUMUS IN SOIL COLUMNS OF OTHER VALLEYS

COACHELLA VALLEY RIVERSIDE COUNTY COACHELLA				OWENS VALLEY INYO COUNTY BISHOP			
Ft.	Soil Clay 16.90	Humus- Nitrogen in		Soil Clay 6.28	Humus- Nitrogen in		
		Humus	Humus Soil		Humus	Humus Soil	
1	Loam.....	.32	5.27 .01	Sandy .....	.31	3.55 .01	
2	Loam.....	.22	5.74 .01	Sandy .....	.23	.....	
3	Loam.....	.14	6.02 .01	Sandy .....	.19	.....	
4	Loam.....	.13	1.08 tr.	Sandy .....	.19	.....	
5	Loam.....	.10	1.40 tr.	Sandy .....	.13	.....	
6	Loam.....	.14	4.01 .01	Sandy .....	.07	.....	
7	Loam.....	.11	5.11 .01	Sandy .....	.08	.....	
8	Loam.....	.14	4.01 .01	Sandy .....	.08	.....	
9	Loam.....	.12	4.68 .01	Sandy .....	.08	.....	
10	Loam.....	.15	5.62 .01	Sandy .....	.06	.....	
11	Loam.....	.16	6.14 .01	Sandy .....	.08	.....	
12	Loam.....	.13	4.32 .01	Sandy .....	.06	.....	
Sum of per cents		1.86			1.56		
Average per foot		.15	4.45 .01		.13	.....	
<i>Upper 3 feet:*</i>							
Sum of per cents		.68			.73		
Average per foot		.23	5.68 .01		.24	.....	

\* Range of most annual plant roots.

penetration. It, however, supported a growth of yucca and desert weeds, and on the same mesa, near Hesperia, with presumably the same soil though deeper, there are a few orchards. A glance at the table shows that there is nearly the same percentage of humus in the first foot of each of the columns and very little difference in the total amount in the upper three feet.

The very unequal distribution in the El Centro column is somewhat surprising, for it might readily be supposed that the small amount in the surface foot would indicate that there was scarcely any in the lower depths; whereas we find higher percentages in the fourth, fifth, eighth, and tenth foot respectively than in either of the upper three feet. It is interesting to note that there is more in the lower half of the column than in the upper. The humus of these columns is not quite as rich in nitrogen as are the soils from other parts of the state, and that of the Bishop column is extremely low. The nitrogen of the soil is not more than 0.01 per cent, or about 400 pounds per acre-foot.

## GENERAL SUMMARY

INDIVIDUAL SOIL COLUMNS HAVING THE HIGHEST  
HUMUS CONTENT

In looking over the 109 columns of soil taken from the various agricultural regions of the state, we find that eighteen have each more than 2 per cent of humus in the surface foot, and thirty-two others have more than 1 per cent, the remaining fifty-nine having less than that.

The following twenty-five localities are worthy of special mention, for in their respective columns more than 1 per cent of humus is found in each individual soil layer to depths of three to seven and even nine feet below the surface; they are ranged according to depth.

TABLE 43.—COLUMNS WITH 1 PER CENT OF HUMUS IN THREE OR MORE  
INDIVIDUAL FEET

Soil	Locality	County	Depth
Tule .....	Stockton .....	San Joaquin .....	9 ft.
Loam .....	Arroyo Grande Valley .....	San Luis Obispo.....	7 ft.
Loam .....	Arroyo Grande Seed-farm .....	San Luis Obispo.....	7 ft.
Clay loam .....	Lompoc Seed-farm .....	Santa Barbara .....	7 ft.
Loam .....	Santa Rosa .....	Sonoma .....	6 ft.
Adobe .....	Berkeley .....	Alameda .....	6 ft.
Loam .....	Russian River .....	Sonoma .....	5 ft.
Clay loam .....	Walnut Creek .....	Contra Costa .....	5 ft.
Adobe .....	El Verano .....	Sonoma .....	4 ft.
Loam .....	Yountville .....	Napa .....	4 ft.
Clay .....	Gilroy .....	Santa Clara .....	4 ft.
Loam .....	Chino .....	San Bernardino .....	4 ft.
Loam .....	Klamath (tule) .....	Siskiyou .....	4 ft.
Loam .....	Glenn .....	Glenn .....	3 ft.
Sandy .....	Chico .....	Butte .....	3 ft.
Clay loam .....	Davis .....	Yolo .....	3 ft.
Clay .....	Farmington .....	San Joaquin .....	3 ft.
Loam .....	Newcastle .....	Placer .....	3 ft.
Loam .....	Kenwood .....	Sonoma .....	3 ft.
Adobe .....	San Ramon .....	Contra Costa .....	3 ft.
Clay .....	Santa Clara Seed-farm .....	Santa Clara .....	3 ft.
Loam .....	Pomona Cienega .....	Los Angeles .....	3 ft.
Clay .....	Santa Ana .....	Orange .....	3 ft.
Clay .....	Alturas meadow .....	Modoc .....	3 ft.
Loam .....	Cedarville .....	Modoc .....	3 ft.

A number of other localities very nearly come into the list as their upper two feet have more than 1 per cent and the third falls a little below. It will be noted that these results are not confined to any particular soil type but that all types from sandy to heavy clay adobe are represented in the list.

If the humus of the upper three feet be equally distributed through that depth we will find that nine of the columns have each an average of more than 2 per cent of humus in

TABLE 44.—SOIL COLUMNS HAVING THE HIGHEST HUMUS PERCENTAGES

IN FIRST FOOT		AVERAGE OF UPPER THREE FEET		AVERAGE OF ENTIRE COLUMN OF 10 OR 12 FEET	
	Per cent		Per cent		Per cent
Stockton tule .....	14.10	Stockton tule .....	16.68	Stockton tule .....	6.81
Pomona Cienega .....	6.26	Pomona Cienega .....	3.67	Arroyo Grande .....	1.39
Santa Clara adobe.....	4.43	Santa Clara adobe.....	3.63	Santa Clara (10 feet) ..	1.33
Cedarville .....	4.29	Klamath marsh .....	3.32	Pomona Cienega .....	1.28
Arroyo Grande seed-farm .....	3.78	Cedarville .....	2.25	Arroyo Grande seed-farm .....	1.27
Klamath marsh .....	3.75	Gilroy .....	2.23	Gilroy .....	1.14
Alturas meadows .....	3.05	Arroyo Grande .....	2.16	Cedarville (10 feet)....	1.10
		Arroyo Grande seed-farm .....	2.15	Lompoc seed-farm .....	1.09
Gilroy .....	2.76	Berkeley adobe .....	2.01	Berkeley .....	1.04
Cedarville meadows....	2.71	Yountville .....	1.98	Russian River .....	1.01
Yountville .....	2.64	Cedarville meadows....	1.89	Yountville .....	.95
Lompoc seed-farm .....	2.50	Lompoc seed-farm .....	1.86	Santa Rosa .....	.92
Arroyo Grande .....	2.50	Alturas meadow .....	1.84	El Verano (10 feet)....	1.00
Santa Ana adobe.....	2.42	Kenwood .....	1.74	Walnut Creek .....	.80
Chino moist land .....	2.31	El Verano adobe .....	1.71	Hayward .....	.77
Kenwood .....	2.25	Farmington .....	1.65	Davis Univ. Farm.....	.75
El Verano adobe .....	2.14	Santa Ana adobe.....	1.63	Watsonville .....	.74
Berkeley adobe .....	2.13	Russian River .....	1.58	Chico .....	.73
Farmington .....	2.04	Chino .....	1.52	Vacaville .....	.71
Vacaville .....	1.97	Glenn .....	1.51	Glenn .....	.69
Santa Rosa .....	1.95				

the upper three feet, and twenty-five others have an average of more than 1 per cent, all others falling below the 1 per cent. In the distribution of the humus through the entire twelve feet of the columns there are ten which thus give an average of more than 1 per cent for each foot, and twenty-five others whose average is about one-half of 1 per cent.

In the following table we have placed the names of the twenty localities which have as much as 1.95 per cent in the surface foot, and the twenty which rank highest in averages of the upper three feet and in the entire column of ten or twelve feet.

#### COMPOSITE COLUMNS OF AGRICULTURAL REGIONS

In the following table are given the averages of each of the agricultural regions embracing all of the 109 columns representing the state at large, taken from thirty-seven counties. The tule marshes of Stockton, Pomona, and Klamath are placed in a column by themselves. In some of the individual columns that form the composite for each region either a depth of twelve feet was not secured or the humus itself was not found to that depth.

The marsh lands are naturally richest in humus because of the large amount of decaying vegetable matter they hold. But next to these we find that the Coast Range valleys have the highest amount in the surface foot, the upper three feet, and in the entire column respectively. The lava-bed valley and meadow lands are but little above the Sacramento Valley in amount of humus, while the San Joaquin Valley falls behind Southern California. The "desert" plains naturally are lowest of the eight groups, but it is interesting to note that in the quite even distribution throughout the column there is a larger amount in the lower six feet than is found in the lower six feet of either the San Joaquin Valley or the lava-bed valleys.

In the foothill and lava-bed valleys the upper two feet hold one-half of the total humus; in the desert lands one-half of the humus is distributed through the upper five feet of soil, while in each of the other columns the upper three feet holds one-half of the humus.

The first foot of the desert lands contains but about one-eighth of the total amount of humus of the column; that of the Coast Range valleys and the marshes, a little more than one-fifth; while in the other regions the upper foot holds from one-third to one-fourth of the total amount found in the respective columns.

On glancing at the table the attention is first called to the depth of twelve feet to which humus reaches in all of the com-



TABLE 45.—COMPOSITE AVERAGES OF AGRICULTURAL REGIONS (IN PERCENTAGES)

Depth	Sacramento Valley 18 columns	San Joaquin Valley 23 columns	Coast Range Valleys 24 columns	Southern California 25 columns	Sierra Nevada Foothills 3 columns	N.E. Lava Beds 8 columns	"Desert" Plains 5 columns	Tule Marshes 8 columns
1st ft. ....	1.04	.80	1.94	.88	1.12	1.55	.26	8.04
2nd ft. ....	.75	.51	1.47	.65	.71	.92	.19	8.59
3rd ft. ....	.58	.37	1.13	.45	.57	.48	.17	7.05
4th ft. ....	.45	.25	.93	.37	.39	.40	.25	5.52
5th ft. ....	.36	.23	.77	.31	.18	.21	.18	2.65
6th ft. ....	.32	.17	.67	.27	.14	.18	.13	1.45
7th ft. ....	.23	.14	.59	.25	.18	.13	.13	.78
8th ft. ....	.19	.10	.49	.19	.10	.12	.14	1.64
9th ft. ....	.18	.18	.41	.16	.....	.11	.12	.68
10th ft. ....	.17	.06	.39	.16	.....	.09	.13	.30
11th ft. ....	.16	.06	.27	.13	.....	.03	.15	.21
12th ft. ....	.15	.04	.32	.11	.....	.03	.13	.29
<i>In 12 feet</i>								
Sum of humus per cents .....	4.58	2.91	9.38	3.93	3.39	4.25	1.98	37.20
Nitrogen in humus	5.45	6.53	5.15	5.54	5.64	3.69	4.69	5.63
Nitrogen in soil....	.03	.02	.04	.02	.03	.03	.01	.02
<i>In upper 3 feet*</i>								
Sum of humus per cents .....	2.37	1.68	4.54	1.98	2.40	2.95	.62	23.68
Nitrogen in humus	5.79	6.27	5.13	5.74	5.40	3.79	4.97	5.75
Nitrogen in soil....	.04	.04	.07	.03	.05	.05	.01	.44
<i>In surface foot</i>								
Nitrogen in humus	5.68	6.08	5.43	6.50	5.93	3.53	5.03	6.35
Nitrogen in soil....	.05	.05	.10	.05	.07	.05	.02	.47

\* Range of most annual plant roots.

posite columns except that of the Sierra Nevada foothills, the lava-bed meadows and the tule marshes, where, because of the underlying country-rock on the one hand and of water on the other, the depths are limited to eight, ten and seven feet respectively. Seventy-five of the columns have a depth of twelve feet, eleven others a depth of ten feet, and eight a depth of nine feet. In some of the columns the amount of humus is so high in the twelfth foot, especially in that of the valleys of the Coast Range, as to leave no doubt of its being found at a greater depth had

the examination been made. In fact, the column of Fort Romie was carried through fifteen feet, and 0.41 per cent of humus with 6.83 per cent of nitrogen was found in the lowest foot.

The next point of interest is the small amount of humus in the first foot of each of the columns excepting those of the meadowlands and the tule marshes. This is especially noticeable in the composite of the San Joaquin Valley and in that of the "desert" plains. The general average of the surface foot for all of the columns, excluding the marshes, is 1.16 per cent.

Another point of special interest is the gradual diminution of humus percentage downward in each of the composite columns with an occasional slight increase, as is seen in the eleventh foot of the Sacramento Valley and in the twelfth foot of the Coast Range valleys. This decrease indicates smaller amounts of humus-forming vegetable material, presumably the roots of plants, in each successive foot downwards, the main mass of the root systems being in the upper three or four feet.

*Averages of Three Upper Feet.*—A depth of one foot in California does not in reality represent the soil which is at least three feet deep, and it would not be correct or fair to the cultural possibilities of the land to draw conclusions from the humus of the first foot alone. It is very true that its presence to the extent of several per cent near the surface is of special importance in maintaining proper physical textural conditions for aeration, avoidance of crusts and easy penetration of water, but it is of as great importance that there should be several per cent of humus in each of the upper several feet; for in arid regions it is below the first foot and away from hot and dry soil that the feeding roots of plants prefer to carry on their activities, and it is in the upper three feet that the main mass of fine feeding roots are usually located, and where they must secure the needed plant food supplied by the humus. This not only protects the roots but gives to them a far greater feeding area which is enlarged with the extension of the humus downward. The summations for three feet are given at the foot of the table.

*Sacramento Valley.*—This valley, represented in the above table by a composite column of eighteen individual columns, is not only richer than the San Joaquin Valley in the first foot in

humus, but in every foot of the entire column to a depth of twelve feet. One half of the total amount is held in the upper three feet. Its first foot contains a little less than that of the Southern California column, but in the upper three feet and in the entire column there is more. The percentage in the first foot is too small and clearly shows the need of being increased by a system of growing and turning under of green crops. With this to encourage the growth of the young grain and trees, the lower portion of the column will afford humus-nitrogen and other plant food for the roots that find their way downward to twelve or more feet as was the case with wheat and barley roots on the University Farm at Davis. In the latter soil the humus of the first foot was only 0.85 per cent and that of the second foot 1.49 per cent, and the ordinary yield had been but from twelve to fourteen bushels of wheat per acre before the University bought the property. By proper methods of treatment and without irrigation or fertilization the agronomist in charge, Professor G. W. Shaw, secured a yield of 40.4 bushels of wheat per acre as an average of three years, during which time the average for the state was but 14.5 bushels.<sup>7</sup>

The humus-nitrogen content of the upper three feet of the Sacramento composite column is 5.79 per cent of the humus which thus gives 0.04 per cent to the soil. This is approximately 1600 pounds of nitrogen per acre for each foot in depth.

*San Joaquin Valley.*—The composite of twenty-three columns from the San Joaquin Valley shows humus percentages much below those of other regions, both in the surface foot (0.80 per cent) and in each foot of the entire column. The sum of 3.11 per cent, if contained in the upper foot of the three feet, would be a fair amount, though much below that of other regions. The low percentages may be due in part to the presence of alkali salts in some of the soils examined, also to a more arid climate of far less rainfall than in other regions, and to a less luxuriant vegetation upon the decay of whose roots the amount of humus is dependent.

Although this humus percentage is so low in the upper part of the soil, yet a proper system of deep plowing to break up any

<sup>7</sup> Bull. no. 211, Cal. Agr. Expt. Sta.

tendency to form a plowsole and to make the soil loose and of good texture for a downward development of the grain roots, which may thus secure the benefits of the humus, would tend greatly to increase the grain yields of the valley. This was shown in the experiments of Professor G. W. Shaw at Ceres and Tulare,<sup>\*</sup> where during a period of three years by this method the average yield of wheat was thirty-five and thirty-three bushels respectively, during which period the average wheat for the state was 14.5 bushes per acre. No irrigation or fertilization was used in his experiments.

The humus-nitrogen of the upper three feet of the composite column is 6.27 per cent of the humus, which is higher than in any of the other composite columns in the table. In the soil itself, however, there is but an average of 0.04 per cent, or 1600 pounds per acre-foot.

*Sierra Nevada Foothills.*—The composite of three columns from the Sierra Nevada foothills shows more humus in the first foot than in that of the Sacramento Valley or Southern California. That of the second foot is, however, somewhat less, and there is a greater decrease in the lower part of the column, the sum being but 3.39 per cent in the eight feet. More than one-half of the humus is held in the upper three feet.

The humus-nitrogen in the upper three feet of the composite column is 5.4 per cent of the humus, or 0.05 per cent in the soil, and is equivalent to 2000 pounds for each acre-foot in the three feet.

*Coast Range Valleys.*—The composite column of the twenty-four soils of the valleys of the Coast Range shows a general average of nearly 2 per cent of humus in the surface foot and thus is far above each of the other columns, except the marshes and meadowlands. Not only that, but it is the only composite column of the uplands which has more than 1 per cent in the subsoils immediately below the first foot. The percentage of humus in each of the succeeding feet throughout the twelve feet is also higher than in any other of the composite columns; the twelfth foot has nearly one-half of 1 per cent and the sum of the entire column is 9.38 per cent. The humus is not as rich in nitrogen

<sup>\*</sup> Bull. No. 211, Calif. Agr. Exp. Sta.

as that of the San Joaquin or Southern California, and the amount of organic nitrogen in the soil is below the 0.05 per cent regarded as necessary for fertility.

*Southern California.*—The soils of the valleys of Southern California represented by the composite of twenty-five columns have about the same humus content in the first foot and in the upper three feet as was found in similar parts of the columns from the Sacramento Valley and the foothills, but are far behind those of the Coast Range valleys. The humus is well distributed downward to twelve feet, and as with the Sacramento soils affords an excellent and large feeding area for plant roots; every encouragement should be given crops to take advantage of this by sending their roots deep into this fertile soil mass. The upper three feet contains a little less than one-half of the total humus.

The humus of the first foot is too low for the maintenance of good physical condition and careful attention should, as a rule, be given to the turning under of green crops and the humification of the same.

The humus in the upper three feet contains 5.74 per cent of nitrogen, but this gives only the small amount of 0.03 per cent to the soil: this is equivalent to about 1200 pounds per acre-foot, which under the influence of bacteria gradually becomes available for plants.

*"Desert" Plains.*—The desert lands represented by a composite of five columns from the valleys of Imperial, Coachella, Owens River, and Mojave River are very low in humus in their upper several feet, as is to be expected from the meagerness of the natural humus-forming vegetation. One-half of the humus is held in the upper five feet instead of in three, as is the case with other regions. The presence of so much humus in the twelfth foot is a matter of some surprise, for the lands seem usually quite deficient in natural moisture other than hygroscopic for hundreds of feet in depth. The amount of humus in the twelfth foot is one-half that of the first foot. The humus of the upper three feet contains but 4.97 per cent of nitrogen, which gives but 0.01 per cent of organic nitrogen in the soil: this is equivalent to about four hundred pounds per acre-foot, which is very little.

*Lava-bed Region.*—The valley lands of the Lava-bed region

of the northeastern part of the state have in the composite of eight columns a high percentage of humus, not only in the first foot but in the second. This is natural as the soils are moist from underlying water and there is a strong vegetation whose roots penetrate deeply. The total sum of humus percentages in the twelve feet is 4.25 per cent, the upper three feet holding more than one-half of it. The humus is poorer in nitrogen than in any of the regions except the desert, the average in the upper three feet being 3.79 per cent, or 0.05 per cent in the soil.

*Tule Marshes.*—The tule marshes near Stockton and Klamath and the Pomona cienega have extremely high humus percentages in each of the four upper feet, and the humus is also quite high through the column of seven feet, all derived from the large amount of decaying tule roots. The humus-nitrogen of the upper three feet is, however, only 5.75 per cent in the humus or 0.44 per cent in the soil; the latter is much higher than found in any of the columns.

#### NITROGEN IN THE HUMUS AND IN THE SOIL

Nitrogen exists in the soil partly in the free state in the air that permeates the soil; partly in the vegetable and animal material that has not undergone humification; partly in the humified vegetable and animal matter; and partly as nitrates soluble in water, and hence very variable in amount from day to day and liable to be lost by drainage. That the unhumified material in the soil does not yield its nitrogen to the plants until after complete humification has been shown by the experiments of Professor Hilgard, whose conclusions are as follows:

“It thus appears that although the nitrogen of the unhumified organic matter constituted about 40 per cent of the total in the original soil, it would during the entire year have contributed only to an insignificant extent to the available nitrate supply; while the fully humified ‘matiere noire’ contributed fourteen times as much. During the growing-season of four or five months the unhumified organic matter would have yielded practically nothing to the crop.”<sup>9</sup>

<sup>9</sup> Soils (Macmillan & Co., 1906), p. 360.

The humus itself then is the most reliable source of nitrogen, keeping it in reserve to be given to the roots of plants by degrees by ammonifying and nitrifying bacteria and at the time when most needed, namely, in the growing season. The fertilizing value of humus depends, as has already been remarked, upon the amount of nitrogen that it contains and which may be changed to ammonia and nitrates through the agency of bacteria and given to the soil and plant. The nitrogen content naturally varies according to the nature of the materials from which the humus was formed and to any diminution that may have occurred from bacterial action or other causes, and we therefore find great differences in one and the same column of soil. Sometimes the difference is very great between one foot and the next, for which it is difficult to account.

*Nitrogen in Humus.*—More than one thousand of these nitrogen determinations in the humus of the soil columns have been made and the results were found to vary from 1 per cent to 24 per cent, but the greater number were from 4 per cent to 6 per cent.

The highest percentage of nitrogen in humus was 24.10 per cent, found in the eighth foot of the Berkeley adobe column, and in this were three other levels with more than 10 per cent, all of which were below the sixth foot. The column from Kenwood, Sonoma County, had in its sixth foot 23.2 per cent, while two others of its series had above 10 per cent. The sixth foot of the Santa Paula soil had 20.5 per cent and in the fifth and ninth foot there was more than 10 per cent. The humus of the entire soil column from Fullerton, from the place of Dr. S. S. Twombly, has the credit of being richer in nitrogen than any soil thus far examined, the fifth foot having 20.5 per cent, the eighth and tenth having more than 19 per cent each, the third 18.4, and the others above 10 per cent, excepting the second foot which has 7.8 per cent; the general average of the column is 15 per cent. The amount of humus is, however, quite small throughout the column, and hence the amount of humus-nitrogen given to the soil is but 0.05 per cent. The humus of the Kearney Park soil is another notable example of rather high percentages of nitrogen, as in each of the seven feet below the

upper foot there is more than 10 per cent; but here again the amount of humus is small. The Chico column should also be mentioned, as from the fifth to the twelfth the humus of each foot contains from 10 to 14 per cent of nitrogen, except the ninth which has 9.5 per cent.

In the one thousand determinations of nitrogen there were but sixty-four instances, or about 6.4 per cent, where the humus contained more than 10 per cent of nitrogen: fourteen of these have from 15 to 20 per cent and six have above 20 per cent. The general average of all, including the marshes, is 5.92 per cent for the first foot, 5.60 per cent for the upper three feet and 5.57 per cent for the entire depth of twelve feet.

The humus of the surface foot of the composite of Southern California soils is richer in nitrogen than that of any of the other regions, though nearly equalled by that of the tule marshes. That of the lava-bed valleys is poorer in nitrogen than any.

When we consider the upper three feet of the composite columns we find that the humus of the San Joaquin Valley is much the richest and that of the lava beds again the poorest. Four of the eight columns fall below the general average for the state. The same is true for the averages of the twelve foot columns.

The differences in the percentages of nitrogen in the humus of the upper three feet and of the total column are not very great, especially in the valleys of the Coast Range, where the two are very nearly the same; in the Sacramento Valley, Southern California lava beds and the desert plains the average in the column is considerably less.

*Organic Nitrogen in the Soils.*—It is well to remark here that 0.10 per cent of humus-nitrogen in the upper foot of a humid soil is regarded as an ample supply for fertility, while in the arid region with its deeper soil, deeper humus and deeper root penetration, one-half of that amount, or 0.05 per cent in each of the three upper feet is considered sufficient for many years, because the roots are in a deeper feeding area than in the humid region, and bacterial activity is greater and deeper. This percentage would mean 2000 pounds of nitrogen per acre in each foot.



The marsh or tule soils of the state have very high amounts of nitrogen, as shown in the Stockton tule (0.83 per cent), Klamath tule (0.34 per cent), and the Pomona cienega (0.25 per cent). The nitrogen is, however, less available than that of the dry uplands, and a comparison cannot be made. The two soils from the Arroyo Grande Valley, including the Routzahn seed-farm, are highest among the other soil columns with their 0.16 per cent of nitrogen each; the next highest are Gilroy with 0.15 per cent; Berkeley and Farmington each with 0.14 per cent; Burpee seed-farm of Lompoc, Vaca Valley, and Kenwood each with 0.13 per cent; and El Verano, Santa Maria, Chico, Yountville, and Chino each with 0.11 per cent. Newcastle soil has 0.10 per cent, while all others have less than that. The results show most certainly that the lands represented by these fourteen soils should not need fertilization with nitrate fertilizers for many years.

Nine of the soils have but 0.01 per cent of humus-nitrogen in the first foot, seven have but 0.02; forty-six or nearly half of the columns have less than 0.05 per cent, and hence are much below the desirable percentage of nitrogen. The average of the first foot of all of the columns, except the marshes, is about 0.05 per cent.

Some of the upland soils are quite rich in organic nitrogen in the upper three feet, the true soil of the arid region. The adobe of Berkeley is the richest with its 0.13 per cent or approximately 15,000 pounds per acre in a depth of three feet. Eight columns have averages of 0.10 and upward, viz., the two Arroyo Grande soils, Gilroy, Chico, Farmington, Kenwood, Newcastle, and Cottonwood. Thirty-one columns have averages of from 0.06 to 0.10 per cent, all others being below this amount. In other words we find that more than half of the columns have less than the 0.05 per cent of organic nitrogen in their upper three feet which is considered necessary for fertility. Many have but little more than traces of nitrogen.

In the entire column of twelve feet we find that there are eleven upland localities that have averages of as much as 0.05 per cent, and among these Berkeley and Arroyo Grande columns stand highest with averages of 0.08 per cent or approximately

38,000 pounds of organic nitrogen distributed through the depth of twelve feet.

#### HUMUS IN SOILS OF DIFFERENT TEXTURE

We have grouped together in the following table the humus percentages of soil columns having similar texture and similar characters in order to ascertain whether these have any influence upon the amounts of humus. For the textural classification of the soils we have used that of Professor Hilgard,<sup>10</sup> the basis of which is the percentage of clay in the soil. Thus soils with 3 per cent and less are classed as very sandy, those with from 3 to 10 per cent of clay as sandy, with from 10 to 15 per cent as sandy loams, with from 15 to 25 per cent as clay loams, with 25 to 35 per cent of clay as clay soils, and those with more than 35 per cent of clay are classed as heavy clays, among which are the adobes of California and other states of the arid west.

TABLE 46.—AVERAGE OF HUMUS IN LANDS OF DIFFERENT TEXTURE AT VARIOUS DEPTHS

	Very sandy 1-3% clay 9 columns Per cent	Sandy 3-10% clay 15 columns Per cent	Sandy loam 10-15% clay 21 columns Per cent	Clay loam 15-25% clay 23 columns Per cent	Clay 25-35% clay 8 columns Per cent	Heavy adobe 35% clay 10 columns Per cent
1 ft.....	.62	.81	.97	1.33	2.03	1.56
2 ft.....	.50	.62	.77	.98	1.25	1.23
3 ft.....	.37	.43	.55	.73	1.11	.95
4 ft.....	.31	.35	.43	.63	.89	.61
5 ft.....	.29	.35	.37	.48	.68	.43
6 ft.....	.21	.35	.31	.41	.52	.37
7 ft.....	.20	.31	.30	.34	.53	.28
8 ft.....	.14	.26	.24	.28	.40	.21
9 ft.....	.12	.20	.19	.24	.37	.17
10 ft.....	.07	.19	.20	.22	.29	.17
11 ft.....	.04	.15	.14	.20	.19	.14
12 ft.....	.03	.16	.12	.19	.25	.13
<i>Sum of per cents:</i>						
Upper 3 feet....	1.49	1.86	2.29	3.04	4.39	3.74
Upper 6 feet....	2.30	2.91	3.40	4.56	6.48	5.15
Lower 6 feet....	.60	1.27	1.19	1.47	2.03	1.10
Entire column..	2.90	4.18	4.59	6.03	8.51	6.25

<sup>10</sup> Soils (Macmillan & Co.), p. 84.

It is interesting to note in each of these composite columns of soils the quite regular decrease in the amount of humus from the surface to the twelfth foot, thus indicating that in each the humus has been derived from the decay of plant roots rather than from vegetable material deposited during the building-up of the soil strata. It also gives testimony to the deep rooting of plants of all kinds, notably grasses and weeds during the ages that have passed.

Another interesting point is the regular increase in the humus percentages from a minimum in the very sandy to the maximum in the clay and then a decrease in the adobe clays, not only in the surface foot of these composite soil columns but also in each succeeding foot down to the twelfth, the only exceptions being slight ones in the sandy loam series which are a little less than in the sandy.

It is clearly evident, then, that as a rule the clay soils, excepting the adobe clays, of the state have a higher percentage of humus than any other class; and that the humus content in the series increases with the increase in clay up to a certain point when there is a decrease.

The tables show that the lower half of the heavy adobe clays has a smaller percentage of humus than any other class except the very sandy; the inference being that the close, compact, heavy clay strata have prevented the extension of plant root systems to the lighter soils.

It will also be noted that one-half of the total humus in each of the composite columns is held in the upper three feet, with the exception of the sandy, in which it is held in four feet.

#### COMPARISON OF INDIVIDUAL HEAVY BLACK ADOBE CLAY SOILS

The heavy clay soils known as adobe clay and containing from 35 per cent of clay upward to as high as 62 per cent are found in isolated areas throughout the state, the largest being in the region of Stockton. They form a distinct class possessing peculiar physical properties. The surface soil is usually black to a depth of three or more feet, changing to a lighter color below and generally resistant to the easy movement of water, plant roots, and tillage implements, though the roots of ordinary

weeds have been followed to the depth of eleven feet below the surface, and alfalfa roots have been observed at a depth of twenty-six feet in the adobe of Berkeley. It is extremely plastic when wet, but when dry it absorbs water with extreme slowness. Because of the black color it was supposed that the humus content would be found to be quite high in all localities, but the disappointment following the examination of many columns has induced us to present them in a table for comparison; the figures for humus and for amount of clay are taken from the report on the several agricultural regions already given. The "adobe" soils in which there is less than 35 per cent of clay are omitted from the table.

TABLE 47.—PERCENTAGE OF HUMUS IN COLUMNS OF BLACK ADOBE CLAY

Depth	Santa Clara 59% clay	Berkeley 44% clay	East of Willows 48% clay	Walnut Creek 35% clay	San Ramon 45% clay	South of Yuba City 39% clay	Stockton 57% clay	Kings City 35% clay	South of Dixon 62% clay	West of Tracy 40% clay	Biggs 50% clay
1 ft.	4.43	2.13	1.72	1.42	1.23	1.20	1.16	1.15	.86	.82	.66
2 ft.	3.66	2.04	1.16	1.44	1.28	.21	.76	.90	.62	.70	.49
3 ft.	2.80	1.84	.94	1.16	1.08	.18	.50	.61	.34	.30	.40
4 ft.	.61	1.90	.62	1.12	.84	.18	.22	.25	.28	.12	.17
5 ft.	.27	.95	.36	1.08	.81	.11	.22	.52	.30	.19	.06
6 ft.	.20	1.06	.26	.72	.75	.10	.41	.41	.30	.09	.06
7 ft.	.62	.48	.22	.60	.29	.08	.11	.43	.30	.....	.09
8 ft.	.23	.37	.20	.60	.20	.12	.09	.37	.26	.00	.06
9 ft.	.30	.36	.14	.42	.14	.12	.08	.33	.07	.00	.05
10 ft.	.13	.36	.22	.52	.12	.12	.07	.11	.07	.00	.04
11 ft.	.....	.45	.20	.36	.09	.13	.09	.....	.09	.00	.06
12 ft.	.....	.49	.16	.26	.07	.....	.11	.....	.09	.00	.09

It is a matter of much surprise that the heavy black clay or adobe soils of the state should, generally, have so little of humus in the upper three feet and especially in the surface soil. Only two of the eleven columns chosen as typical of the various black clay regions have as much as 2 per cent in the upper foot. The black adobe of the Sacramento and San Joaquin valleys have far less humus than was found in those of the Coast Range. It is also a surprising fact, well shown in these soils, that a black soil does not always owe its color to the humus that it contains, for all of those given in the table are very black and yet have but little humus; this is especially noticeable in those from

Tracy, Biggs, and south of Dixon, which have less than 1 per cent. A very striking example of this was found in a sample of adobe soil sent for examination some time ago from near the hills in the Santa Clara Valley, which although almost jet black had but a trace of humus.

It is further interesting to note that in the Berkeley and Walnut Creek columns there is more than 1 per cent of humus in each of the upper six and five feet below the surface respectively; in each of the upper three feet of the Santa Clara and San Ramon columns and in the upper two feet of the Willows column. In the Dixon, Tracy, and Biggs columns there is less than 1 per cent in the first foot.

In the fourth foot of the Santa Clara, seventh foot of the Berkeley, and third foot of the Yuba City there are very sharp drops in the percentages, and this is seen also in the lighter color of the respective columns at those depths. These indicate that the main root systems of the plants were confined to the upper part of these soils, while in other columns there was less of root mass and a greater downward development. In all except where ground water was found above the twelfth foot, the humus clearly could be found at greater depth, especially in the Berkeley column, which had 0.49 per cent in the twelfth foot.

The Santa Clara soil has in its first foot the highest percentage of humus of any soil of the state thus far examined, excepting the tule marshes; the same is true if we compare the average of the three upper feet and also that of the upper six feet, which are respectively 3.62 and 1.99 per cent. The general average of humus in the first foot of the eleven columns is 1.61, for the second 1.20, and for the third foot only 0.92 per cent.

#### RELATION OF HUMUS PERCENTAGE TO COLOR AND OTHER SOIL CHARACTERS

There are types of land having other than textural characteristics concerning whose relation to humus content some question may arise, and we have segregated some of the more important of these in the following table. They refer to the uplands and lowlands, the several color characters and to strong

alkali lands. We have omitted the tule marsh lands from the composite of lowlands, and the strong alkali lands and the desert lands from the composite of uplands.

TABLE 48.—AVERAGE HUMUS IN SOILS OF DIFFERENT CHARACTER AND COLOR

	Stream alluvial 5 columns	Non-alkali uplands 72 columns	Black lands 17 columns	Grav lands 16 columns	Red lands 10 columns	Alkali land 4 columns
1 ft.....	1.41	1.18	1.19	.94	.64	.30
2 ft.....	1.09	.91	.45	.69	.42	.26
3 ft.....	.84	.65	.38	.57	.28	.23
4 ft.....	.76	.37	.26	.47	.24	.18
5 ft.....	.68	.42	.22	.44	.24	.16
6 ft.....	.56	.36	.21	.40	.17	.08
7 ft.....	.52	.35	.15	.37	.12	.08
8 ft.....	.41	.27	.12	.30	.10	.06
9 ft.....	.34	.25	.11	.31	.11	.04
10 ft.....	.24	.24	.09	.28	.11	.04
11 ft.....	.26	.23	.05	.25	.12	.04
12 ft.....	.23	.23	.08	.22	.18	.03

## COMBINATION OF UPPER AND LOWER PORTIONS

*Sum of per cents:*

Upper 3 feet....	3.34	2.74	2.02	2.20	1.34	.79
Upper 6 feet....	5.34	3.89	2.71	3.51	1.99	1.21
Lower 6 feet....	2.00	1.57	.60	1.73	.74	.29
Total 12 feet....	7.34	5.46	3.31	5.24	2.73	1.50
Average per ft.	.61	.45	.28	.44	.23	.12

The table shows the interesting and unexpected fact that in their general averages the alluvial lands that border the streams are but little richer in humus than the uplands, either in the surface foot or at the several levels to the twelfth foot. In the fourth foot the difference is 0.39 per cent, but in every other level it is less than that. In the upper three feet the difference is 0.60 per cent, in the upper six feet 1.45, and for the entire column it is only 1.88 per cent in favor of the alluvial lands. There are, of course, throughout the state instances of high percentages in single soils of both the alluvial and upland classes, and but for these the general averages would be much lower.

The close agreement in humus percentages of the uplands and alluvial lands, and the gradual and quite uniform diminution of the same downward plainly indicates that the source of the

humus in both classes is in the decay and humification of plants whose roots had penetrated to depths of many feet.

The small differences throughout the composite columns would also show that the alluvial lands and the deltas possess no material advantage, except perhaps in moisture conditions, over the higher lands which are of good depth and free from hardpan.

*Relation of Humus to Soil Color.*—It has already been pointed out that a black color does not necessarily mean the presence of a high percentage of humus, and this is again emphasized in the composite columns of many soils of three colors taken from the main agricultural regions of the state. In these the surface foot of the black has but little more humus than that of the gray soil (1.19 and 0.94 per cent respectively). But still more surprising is the fact that, although the upper three feet of the black lands are almost invariably very dark and even black, they contain less humus than the upper three feet of the gray sandy lands; and that throughout the entire composite columns the percentage of humus is greater in the gray than in the black soils with the exception of the first foot. There are, of course, black clay lands of Santa Clara, Arroyo Grande, and other localities which have high percentages, but these are offset by those with very small amounts.

The composite of eleven red lands embracing mesa and hill lands and the more level lands on the eastern side of the great valley contains less humus than either the black or the gray, except in the lower six feet.

\*The gray lands of the state seem therefore to have greatly the advantage over both the black and red classes in larger humus content and in its better distribution throughout the depth of twelve feet.

*Humus in Strong Alkali Soils.*—This column is a composite of four alkali soils from the San Joaquin Valley, including the Tulare Lake bed, and shows a deficiency in humus throughout. Alkali salts are usually held in the upper four feet of these valley soils and it is there where they would prevent the growth of vegetation with accompanying root systems whose decay would result in the production of humus. Hence we find but little more than mere traces of humus below the fifth foot.

## HUMIC PHOSPHORIC ACID IN SOILS

While one of the chief values of humus lies in its content of nitrogen which is made available to plants by bacterial action, the mineral elements potash and phosphoric acid held by it are also of value to plants as fertilizers and are probably immediately available. The amount of phosphoric acid is especially quite large, as is shown by an analysis of the ash from humus of a productive prairie soil of Minnesota made by Professor Snyder.<sup>11</sup>

TABLE 49.—ANALYSIS OF THE ASH OF HUMUS

	Per cent
Ash (precipitated humus) .....	12.24
COMPOSITION OF THE ASH	
Silica .....	61.97
Potash .....	7.20
Soda .....	8.13
Lime .....	0.09
Magnesia .....	0.36
Ferric oxid .....	3.12
Alumina .....	3.48
Phosphoric acid .....	12.37
Sulfuric acid .....	0.98
Carbonic acid .....	1.64

Early in the investigation of the humus in these soil columns, we made examinations of the ash of the humus of a few of the soil columns to their full depth to ascertain how much, if any, phosphoric acid was present in an organic form combined with the humus below the upper few feet. The difficulty of securing a humus solution free from suspended clay makes the results given in the following table only approximately correct, though sufficiently so to be of special interest in showing that phosphoric acid does accompany the humus in all soils to depths of many feet, and being thus distributed downward throughout the soil mass is at the command of roots as they reach to water.

<sup>11</sup> Chemistry of Soils and Fertilizers.



TABLE 50.—PERCENTAGE OF HUMUS AND HUMIC PHOSPHORIC ACID IN SEVERAL OF THE SOIL COLUMNS

	Orchard Yuba City Clay loam		Orchard Chico Alluvial		Grain field Stockton Adobe clay		Grain field Modesto Sandy		Hop field Russian River Alluvial		Seed farm Santa Clara Adobe clay		Orchard Santa Paula Clay loam	
	Humus	Phosphoric acid	Humus	Phosphoric acid	Humus	Phosphoric acid	Humus	Phosphoric acid	Humus	Phosphoric acid	Humus	Phosphoric acid	Humus	Phosphoric acid
1 ft.....	1.27	.05	1.84	.05	1.16	.05	.37	.01	1.80	.07	4.43	.07	1.23	.07
2 ft.....	1.08	.04	1.18	.04	.76	.03	.27	.01	1.24	.09	3.66	.04	1.36	.07
3 ft.....	.45	.02	1.16	.03	.50	.02	.19	.01	1.20	.08	2.80	.02	.52	.04
4 ft.....	.39	.02	.88	.02	.22	.02	.18	.02	1.18	.06	.61	.06	.57	.04
5 ft.....	.67	.02	.58	.01	.22	.04	.17	.01	.75	.03	.27	.05	.54	.04
6 ft.....	.48	.03	.42	.02	.18	.03	.16	.01	.61	.02	.20	.05	.48	.02
7 ft.....	.22	.03	.40	.01	.11	.02	.17	.01	.48	.03	.62	.09	.54	.04
8 ft.....	.16	.02	.36	.01	.09	.01	.....	.....	.78	.07	.23	.05	.45	.05
9 ft.....	.12	.02	.38	.02	.08	.02	.....	.....	.64	.07	.30	.08	.36	.05
10 ft.....	.11	.01	.46	.02	.07	.01	.....	.....	.52	.06	.....	.....	.51	.04
11 ft....	.12	.01	.32	.02	.09	.02	.....	.....	.54	.04	.....	.....	.60	.03
12 ft.....	.11	.01	.52	.02	.11	.02	.....	.....	.22	.04	.....	.....	.52	.03
<i>Total available phosphoric acid in soil:</i>														
1st ft.	.18		.09		.06		.11		.18		.12		.12	
6th ft.	.11		.10		.11		.11		.23		.10		.19	
12th ft.	.09		.13		.23		.13		.26		.12		.22	

It will be seen from this table that the highest amounts of phosphoric acid in organic combinations occur in the upper two feet of each column with the exception of the Russian River and the Santa Clara, each of which have high percentages in their lowest depths, although the amount of humus is much lower than in the upper part.

The general average is 0.05 per cent for each of the two upper feet of the seven columns, 0.03 per cent for each of the succeeding eight feet, and 0.02 per cent for each of the two lowest feet of the five columns whose depth reaches twelve feet respectively.

If we estimate the percentages into pounds per acre, we find that the amount of this humic phosphoric acid for the column is approximately as follows:

	Surface foot	Upper 3 feet	Entire column
Yuba City, Sutter County .....	2,000 lbs.	4,800 lbs.	11,200 lbs.
Chico, Butte County .....	2,000 lbs.	4,800 lbs.	10,800 lbs.
Stockton, San Joaquin County .....	2,000 lbs.	3,600 lbs.	11,600 lbs.
Russian River, Sonoma County .....	2,800 lbs.	9,600 lbs.	26,400 lbs.
Santa Paula, Ventura County .....	2,800 lbs.	7,200 lbs.	15,600 lbs.
Santa Clara, Santa Clara County....	2,800 lbs.	4,800 lbs.	20,400 lbs.
Modesto, Stanislaus County .....	400 lbs.	1,200 lbs.	3,200 lbs.

Even in the upper foot of each of the above there is from 2000 to 2800 pounds, with the exception of that of Modesto, which has only 400 pounds of phosphoric acid per acre.

When we consider the fact that 0.05 per cent or 2000 pounds per acre of available phosphoric acid is regarded as a sufficient amount for soil fertility for present needs, we can readily see that these particular soils, except that from Modesto, are abundantly supplied with phosphoric acid for many years.

Analyses of the first, sixth, and twelfth foot respectively of the columns also show the presence of large percentages of available potash.

#### COMPARISON OF ARID AND HUMID SOILS

A comparison between the soils of the arid and humid regions brings out the very great advantage possessed by the former and especially by those of California over the humid, as shown in the first part of this bulletin. But it is especially marked in the amount and distribution of humus in the soils of the respective region.<sup>12</sup> The general average in 280 soils of the humid portion of the United States is 2.63 per cent, and if we add to this the 5.26 per cent in eleven tropical soils of Hawaii the average is brought to 2.73 per cent, or approximately 109,200 pounds per acre in a depth of twelve inches.

The average of the 331 California soils and forty-four from Idaho, Arizona, and Oregon is 1.28 per cent for the surface foot, or less than one-half that of the humid region. For the first

<sup>12</sup> It has been a rather difficult matter to secure data on humus in the soils east of the Mississippi River for the reason that in their analysis some of the chemists have failed to separate it from the percentage of aggregate "organic matter."

foot of the 109 soil columns including the marsh lands the average is 1.35 per cent.

But the depth of soil in which most of the plant roots are held in the arid region, and in which the humus color is most pronounced is *three feet*, and taking this as the true soil of California we find that there is a sum of 3.17 per cent, or an average of 1.06 for each foot; this is equivalent to about 127,000 pounds per acre. To this should be added the percentages of humus found at lower levels of the soil, as shown in the averages of the 109 columns given in the accompanying table, which would bring the total humus to 233,000 pounds in twelve feet.

TABLE 51.—AVERAGE OF HUMUS IN ARID AND HUMID SOILS

Ft.	ARID SOILS		HUMID SOILS	
	109 California soil columns Per cent	842 soils in United States Per cent	291 soils in United States Per cent	
1.	1.35	1.28	Soil	2.73
2. Soil .....	1.04	?		?
3.	.78			00
4. Subsoil .....	.62			00
5.	.45			00
6.	.36			00
7.	.30			
8.	.26			
9.	.21			
10.	.18			
11.	.14			
12.	.14			
Sum of per cents in 12 ft.	5.83			
Sum of per cents in 3 ft.	3.17			
Nitrogen in humus, 3 ft.....	5.60			
Nitrogen in soil, 3 ft.....	.05			

The amount of humus then in the average of California soils (three feet deep) is greater than in those of the humid region; and for the depth of twelve feet it is more than twice as great.

The determinations of humus-nitrogen in the soils of the humid region have been so few and miscellaneous that it is not advisable to deduce from them an average to represent the whole region.

Arid California with her deep soils of well diffused humus and richness in plant food thus possesses very great advantages over the more humid east where a depth of six to nine inches is very generally accepted as that of a true upland surface soil, containing practically all of the humus and marked by a very sharp change of the black humus color to the gray and yellow subsoil with its inappreciable amount of humus.<sup>13</sup> In the sediment lands of the streams and some of the black prairies the humus is of course found at a greater depth.

## CONCLUSIONS

1. The depth of the soils of California is indefinite because of the presence of humus and available plant food to and beyond twelve feet below the surface. It may be several times twelve feet, being limited only by ground water, country rock or heavy beds of gravel; layers of hardpan at a depth of several feet would limit the depth for small cultures, but may be broken up with dynamite and the depth greatly increased for the extension of tree roots. There are in reality none of those difficultly permeable clay subsoils that characterize humid soils and limit their depth to but a few feet. The sharply defined change of the black humus color to gray at six or nine inches that marks the depth of humid soils is present in some of the heavy clay soils in California at the depth of three feet, but for the most part the change in tint is very gradual downward through many feet. The upper three feet may, however, properly be called the soil, for in that depth the greater part of the feeding roots of plants are found.

2. The soils of California are richer in humus than has generally been supposed; in their depth of three feet (the soil proper) they contain more than in the humid soils, and in the entire columns of twelve feet or more they have double that of the humid soils.

<sup>13</sup> See Hilgard's *Soils*, p. 164, and *Agricultural Science*, 1892, p. 263; King's *The Soil*, p. 29; Hall's *Soils*, p. 45; Minn. Agr. Expt. Sta. Bull. 30, 41, and 65 contain analyses of 121 subsoils, in only sixteen of which is humus reported though found in all of the corresponding surface soils which were taken to depths of nine inches. (Bull. 30, p. 164.)

3. The humus in California soils is usually distributed through depths of twelve or more feet, the highest percentage being in the upper three feet and diminishing downward as shown in each of the 109 soil columns taken from the seven agricultural regions; as much as 41 per cent was found in the fifteenth foot of the Ft. Romie column, and 0.74 per cent in the thirteenth foot of that of Davis and Hayward; water was reached in each case.

4. The surface soils of California have an average of 1.28 per cent of humus as shown in the analysis of several hundred samples taken from the many agricultural regions of the state. This is not sufficient to maintain good tilth in the soil; but as supplemented by that of many feet below often affords a fair supply of nitrogen. The upper three feet of soil proper has an average of 1.06 per cent per foot, or a sum of 3.17 per cent.

5. The tule swamps have the highest percentage of humus because of the mass of decaying roots and other vegetable matter, while the desert plains have the least.

6. Humification is retarded in close compact adobe clays and the amount of humus is less than in lighter loam and sandy soils.

7. The valleys of the Coast Range in the western part of the state have higher percentages of humus in their soils than have any of the other agricultural regions of California, probably because of the greater humidity of the climate of that region and a denser vegetation.

8. The humus of the soils of this state is very generally derived from plant roots instead of from accumulations of vegetable material at various depths as the soil was being built up, thus showing that the native vegetation has for ages been deep-rooted.

9. The black color of a soil is not always due to a high humus content; many black soils have a smaller percentage of humus than soils of a gray color. In one instance a very black clay soil from the Santa Clara Valley contained no humus, its color being due to the presence of a black rock powder.

10. Humus contains nitrogen in combination, the amount depending largely on the source of the humus. The average percentage in the humus of the first foot of the soil columns is

5.92 per cent; that of each of the upper three feet is 5.60, and a little less for the entire twelve feet. It varies from 1 to 20 per cent in individual soil layers.

11. The organic nitrogen in the soil derived from the humus and dependent on the amount of the latter, varies from almost nothing in the lower depths of the soil to as much as 0.13 per cent in the upper three feet. The average of the first foot of the soil columns is 0.07 per cent; for each of the three upper feet, the range of most annual plant roots, it is 0.05 per cent. The investigations of Professor Lipman of this station show that nitrifying bacteria are present and active in California soils to depths of six feet and ammonifying bacteria are present through a depth of twelve feet, thus making available to plants the nitrogen content of the humus to these depths.

12. Humus contains soluble mineral plant food in combination, the phosphoric acid being present in the humus of California soils to the extent of from 0.01 per cent to 0.08 per cent throughout the entire depth to which humus reaches, though usually greatest in the upper few feet.

13. Humus is sometimes less in the first foot than in the second because it is gradually destroyed by cultivation and summer fallowing of the soil, but may be replaced and increased by proper methods of green-manuring, or the turning-under and humification of legumes.

14. Arid soils have an immense advantage over those of the humid region of the United States because of this distribution of humus and its nitrogen, as well as of mineral plant food, through a depth of many feet, as it gives a greater depth of soil and induces a deeper root penetration for plants and trees into a greater feeding area and where there is more moisture. The wonderful endurance of drought on the part of California soils is due to this.

15. The practical value and hence the commercial valuation of land in California cannot be based alone upon the nature of the surface soil and subsoil as in the humid region, but chiefly upon its texture and depth, and upon the freedom with which plant roots are able to penetrate to many feet and secure moisture and the abundant food supply that exists at those depths in all of California soils.



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NEW EXPERIMENTS ON ALKALI SOIL  
TREATMENT

*(Preliminary Report)*

BY

CHARLES B. LIPMAN AND LESLIE T. SHARP

UNIVERSITY OF CALIFORNIA PRESS  
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## NEW EXPERIMENTS ON ALKALI SOIL TREATMENT

*(Preliminary Report)*

BY

CHARLES B. LIPMAN AND LESLIE T. SHARP

The presence of "alkali," denoting water-soluble salts in considerable quantities in soils, has long been one of the problems of soil management in arid regions from the point of view of the practical as well as that of the scientific man. While soluble salts like magnesium and calcium sulphates, and chlorides as well as, in some cases, nitrates of the alkalies and alkali earths and other soluble compounds frequently occur with them, the salts which usually are found to cause injury in alkali soils are sodium chloride, sodium sulphate, and sodium carbonate. It is unnecessary to go into detail here on the specific effects of every one of these salts. Suffice it to say, that in general the damage is caused by these salts through corrosion of living plant substance, through plasmolysis of plant cells owing to injurious osmotic influences, and through effects on the physical, chemical, and bacteriological constitution of the soil which result in poor aeration, poor moisture supply, improperly balanced chemical composition of the soil solution, and in impairment of those bacteriological activities necessary to insure an available supply of plant food substances, particularly of the nitrogen variety.

Alleviative measures in vogue to date in dealing with the alkali problem as it affects crop production have been, in general, of two classes. The first class is that concerned with the removal of alkali salts from the affected soils by flooding, accompanied by underdrainage by natural or tile drain methods. The second

class has been that of rendering innocuous, in part or in whole, the alkali salts without their removal from the soil. In the latter a division may again be made into methods of chemical treatment, such as that against sodium carbonate, or "black alkali," with gypsum and its transformation into the less harmful Glauber salt or sodium sulphate, and methods of heavy irrigation and deep tillage for the purpose of disseminating the salts through a larger internal soil surface, thus rendering less concentrated the soil solution of alkali salts. It may be added in this connection that empirical methods of alkali soil treatment, such as those of Symmonds<sup>1</sup> and Darnell-Smith<sup>2</sup> in applying nitric and sulphuric acids respectively, and those of others employing barnyard manure, have in a few instances been rewarded with good results. Fuller details with reference to these experiments need not be given here, since they will be discussed in the more complete report which will appear when our experiments have matured.

#### BASIS OF THE PRESENT EXPERIMENTS

It will be noted above that ameliorative measures in alkali land treatment have been, with the exception of that of the drainage treatment and that of the Hilgard proposal of gypsum treatment against black alkali, of an empirical nature and not based on established scientific principles. Some measures have indeed been employed without any good reason. It occurred to the writers, therefore, to attack the problem of alkali treatment in soils on the scientific basis of principles established on theoretical or experimental grounds. The latter included, broadly speaking, the principle of antagonism between ions,<sup>3</sup> and those of the behavior of soil colloids and chemical soil constituents in the presence of soluble salts, or on the removal of soluble salts.<sup>4</sup>

<sup>1</sup> Journ. Agr. Gov. New South Wales, vol. 21 (1910), p. 257.

<sup>2</sup> Rept. Govt. Bur. Microbiol. New South Wales, vol. 2, p. 209.

<sup>3</sup> See papers of Osterhout in *University of California Publications in Botany* and those of C. B. Lipman in *Centralblatt für Bakteriologie*, 21<sup>o</sup> Abt.; also paper soon to appear by C. B. Lipman and W. F. Gericke, in *Journal of Agricultural Research*.

<sup>4</sup> In detailed studies carried out by L. T. Sharp, which are soon to be published, many data of a fundamental nature have been obtained, on the importance of the relationship existing between soil colloids and soluble salts, particularly when the latter are leached from a soil.

Since the first-named principle is supported by numerous experiments demonstrating the efficacy of some salts in preventing the toxicity of others to plants and to soil bacteria, we have attempted to approach the production of a more balanced soil solution by treating the affected soil with certain salts. Since likewise the experiments of one of us above cited have demonstrated the harmful effects on the soil colloids of the washing out of salts as well as on the removal of necessary elements in the soil, we have attempted to prevent such harmful effects or to neutralize them by the methods of acid and manure treatment which we have employed. The details of our experiments follow below.

#### THE METHOD OF THE EXPERIMENTS

A large quantity of alkali soil was shipped to the greenhouse from a field south of Kerman in the San Joaquin Valley. This soil, which had been previously analyzed for alkali, was found to contain 0.64 per cent of water-soluble salts which was composed as follows: 0.459 per cent NaCl, 0.098 per cent  $\text{Na}_2\text{SO}_4$ , 0.083 per cent  $\text{Na}_2\text{CO}_3$ . It is understood, of course, that the determinations just given are according to conventional analytical methods made referable to sodium as the base, whereas of course other bases must usually occur as above intimated along with the acids determined. In this case, however, only traces of calcium and magnesium were found. The soil just described has never been cropped and has borne only a sparse vegetation of plants resistant to alkali and drouth. It was distributed in eight-inch earthenware pots in portions of six kilograms each and the pots received the following treatments:

- Nos. 1, 2 and 3— Untreated.
- Nos. 4 and 5— 30.42 gr. each of actual  $\text{H}_2\text{SO}_4$  c.p.
- Nos. 6 and 7— 41.76 gr. of actual  $\text{H}_2\text{SO}_4$  c.p.
- Nos. 8 and 9— 11.02 gr. each of actual  $\text{H}_2\text{SO}_4$  c.p.
- Nos. 10 and 11— 62.08 gr. each of actual  $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$  c.p.
- Nos. 12 and 13— 6.00 gr. each of actual  $\text{CuSO}_4$  c.p. calculated as anhydrous salt.
- Nos. 14 and 15— 30.00 gr. each of actual  $\text{FeSO}_4$  c.p. calculated as anhydrous salt.
- Nos. 16 and 17— 12.00 gr. each of actual  $\text{Na}_2\text{SO}_4$  c.p. (anhydrous).
- Nos. 18 and 19— 300.00 gr. each of actual air-dry barnyard manure.

About three days after these treatments were given, selected barley seeds of the Beldi variety were planted in all the pots to the number of thirteen in each. They were later thinned to four plants for each pot. As nearly as possible, optimum moisture conditions were maintained in all the pots throughout the experiment and caution was observed in irrigation so as to obviate any possibility of percolation and the inevitable leaching of salts which would accompany it.

### RESULTS OF THE EXPERIMENT

The seeds were planted on August 2, 1914, and the crop harvested on January 23, 1915. The following table gives the data regarding yields obtained.

TABLE I  
YIELDS OF BARLEY OBTAINED IN ALKALI SOIL TREATMENT

No. of pot	Yield of tops grams	Yield of roots grams	Yield of grain grams	Increased weight of tops over control avg. grams
1	.8	.5	.00	
2	5.5	1.2	.05	
3	8.0	1.7	.05	
4	16.8	6.9	.80	12.80
5	17.0	3.9	.70	12.90
6	4.3	1.1	.05	-0.45
7	15.2	3.0	.10	10.50
8	15.8	3.3	2.60	13.60
9	14.5	3.6	2.20	11.90
10	6.7	1.1	1.15	3.05
11	8.2	.8	1.20	4.60
12	.2	--	.00	-4.60
13	.2	.1	.00	-4.60
14	5.3	.5	.00	0.50
15	12.2	8.7	.50	7.90
16	1.7	.6	.00	3.10
17	1.7	.6	.00	3.10
18	10.0	5.2	.90	6.10
19	7.4	3.9	.90	3.50

The marked effects produced by some of the treatments of the alkali soils are clearly indicated in the data submitted in the foregoing table. Of the three control pots, only Nos. 2 and 3 are probably representative of the true conditions in the soil. But, even taking these larger values for the yields of barley obtained from the untreated soil, it is very striking to note the beneficial effects of several of the treatments. This is especially noteworthy in the case of the sulphuric acid treatments and particularly at the smallest application of that acid. The yields in pots 4 and 5 and pots 8 and 9 are more than three times as large as those of the average yield of the two control pots, 2 and 3. The discrepancy between the duplicate pots (6 and 7) of the largest sulphuric acid application cannot definitely be accounted for, but it is obvious that several explanations might be offered therefor. Even the gypsum, ferrous sulphate, and barnyard manure treatments were instrumental in improving very materially the producing power of the soil for barley. Evidence is now in hand which will be published later indicating that ameliorative results may be obtained with ferrous sulphate far superior to those indicated in Table 1 by using less of the salt and by obviating the deleterious effects of the ferrous salt by allowing it to become partially oxidized in the soil before the seed is planted. It will be noted further that  $\text{CuSO}_4$  and  $\text{Na}_2\text{SO}_4$  were without effect in a positive direction and appeared even to render the soil a much poorer medium for the growth of barley than it was before treatment.

When we pass from the total yields of dry matter to those of the grain produced, the smallest of the three sulphuric acid applications employed seems to be far and away the best treatment of all tested. In respect to the grain yield again, gypsum, and not the intermediate sulphuric acid treatment, stands second, and the latter and the manure treatment are about even.

So far as the root yields are concerned, the data are too irregular to allow of our arrival at any definite conclusions. They do not appear to follow in a general way the yield of tops, are more consistent and regular in the sulphuric acid treatments, and best developed in the manure treatments. In general the favorable treatments were productive of more fibrous root-

development and the unfavorable treatments, or the untreated soils, of short, thick roots having very few fibrous roots. The two exceptionally large root yields in pot 4 and pot 15 are not capable of satisfactory explanation at this time.

It is not our purpose to explain at this time in detail the causes underlying the positive or negative effects of the various treatments, since such explanations will appear in the more complete reports of the work which are to follow. It may be said here briefly, however, that the  $\text{H}_2\text{SO}_4$  exerted its influence, both in the direction of neutralizing the  $\text{Na}_2\text{CO}_3$  and that of improving the soil's physical condition through its shrinkage of colloids. In smaller measure, likewise, gypsum exerted similar effects and in addition thereto exerted the characteristically strong antagonistic effect to the sodium and acid ions which calcium is known to exert in the plant world. The effects of  $\text{FeSO}_4$  are to be explained in general as are those of  $\text{CaSO}_4$ . The effect of the barnyard manure is probably exerted through the organic colloids produced in its decomposition, which through the enormous surface they possess hold much of the salts or components of the latter in a condition which prevents their ready solution in the soil water. Moreover, the organic colloids render the soil more retentive of moisture, therefore diluting the soil solution, and besides exert the most marked effects of any of the materials used in the improvement of physical conditions in the soil. Many other effects are probably also involved in the manure treatment which, like those above mentioned, cannot be discussed here.

Detailed studies are now being made of a chemical, physical and biological nature to determine, so far as may be, the intimate effects concerned in treatments of the soil which are above outlined. Much material has already been accumulated from these supplementary studies which is of great practical as well as theoretical significance.

No less interesting and striking than the data given in Table 1 is the series of photographs accompanying this paper. These confirm in the appearance of the plant growth what is so clearly shown in the record of yields as above discussed, and the reader's attention is particularly directed to them.

### OTHER EXPERIMENTS

In addition to the experiment above discussed, we have now in progress another pot experiment similar to the first, and several field experiments. In the new pot experiment a different soil, coming from Kearney Park near Fresno, is being employed. This soil contains only about 0.44 per cent total salts based on the dry weight of the soil. The total alkali is differently distributed than in the soil of the foregoing experiment, consisting of 0.18 per cent  $\text{Na}_2\text{CO}_3$ , 0.16 per cent  $\text{NaCl}$  and 0.10 per cent  $\text{Na}_2\text{SO}_4$ . It may be stated, briefly, with respect to this experiment that even more striking results are already manifest than in the foregoing series. Nevertheless, it must be added that the barley plants are only six or seven inches high as yet and any predictions as to the final outcome of the experiment would be premature. It is interesting to note, however, that to date none of the control pots shows any growth worth mentioning and that in the treated pots there is better agreement than in the preceding series so far as duplicate pots are concerned.

Of the field experiments above referred to we have five different locations in the San Joaquin Valley, all in the vicinity of Fresno. These consist of small treated and untreated plots in the open alkali fields. Four of the plot experiments concern the growth of barley, while the fifth concerns alfalfa. It is far too early to predict anything as to the outcome of these field experiments, since the period of growth is just beginning, but we have already made observations on one set of plots which gives hopes for success.

### GENERAL REMARKS<sup>5</sup>

The brief statement above made is here submitted to call attention to some striking results already obtained and to the promise of new ones to which present experiments are pointing.

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<sup>5</sup> Since writing the preliminary statement above given the writers have found that owing to too slight a covering of paraffine on the inner walls of the pots some of the salts were absorbed by the porous clay. This can be of little significance only in connection with our statement however, since the largest amounts of salts remain in the pots which gave the heaviest yields. We only advert to the circumstances here for more completeness of record and for future reference.



Obviously the practical significance of any such experiments in the event of their proving successful would be very great. The vast areas of alkali land in this state which at present are worthless could be made to increase California's wealth enormously if they can be treated so as to make them produce, and particularly if they can be treated cheaply. If our experiments should turn out to be entirely successful, as we now have good reason to think they will, treatment of alkali land as outlined above to make it profitable, should prove, relatively, to all other methods, by far the cheapest method of reclamation.

*Transmitted April 16, 1915.*



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ERRATUM

The plates have been misplaced. Plate 1 has been printed as Plate 3, Plate 2 as Plate 1, and Plate 3 as Plate 2. Plate 4 is correctly placed.

The pots have not been numbered. They are arranged in order from left to right; thus, on Plate 1 (printed as Plate 3) are pots 1 to 5; on Plate 2 (printed as Plate 1) are pots 6 to 10, etc.

EXPLANATION OF PLATES

PLATE 1

Treatment of Pots reading from left to right

Pots 1, 2 and 3. No treatment.

Pots 4 and 5. 30.42 grams each of actual  $H_2SO_4$ .



PLATE 2

Treatment of Pots reading from left to right

Pots 6 and 7. 41.76 grams each of actual  $\text{H}_2\text{SO}_4$ . -

Pots 8 and 9. 11.02 grams each of actual  $\text{H}_2\text{SO}_4$ .

Pot 10. 62.08 grams of  $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$ .



### PLATE 3

Treatment of Pots reading from left to right

Pot 11. 62.08 grams of  $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$ .

Pots 12 and 13. 6.00 grams each of  $\text{CuSO}_4$  as anhydrous salt.

Pots 14 and 15. 30.00 grams each of  $\text{FeSO}_4$  as anhydrous salt.



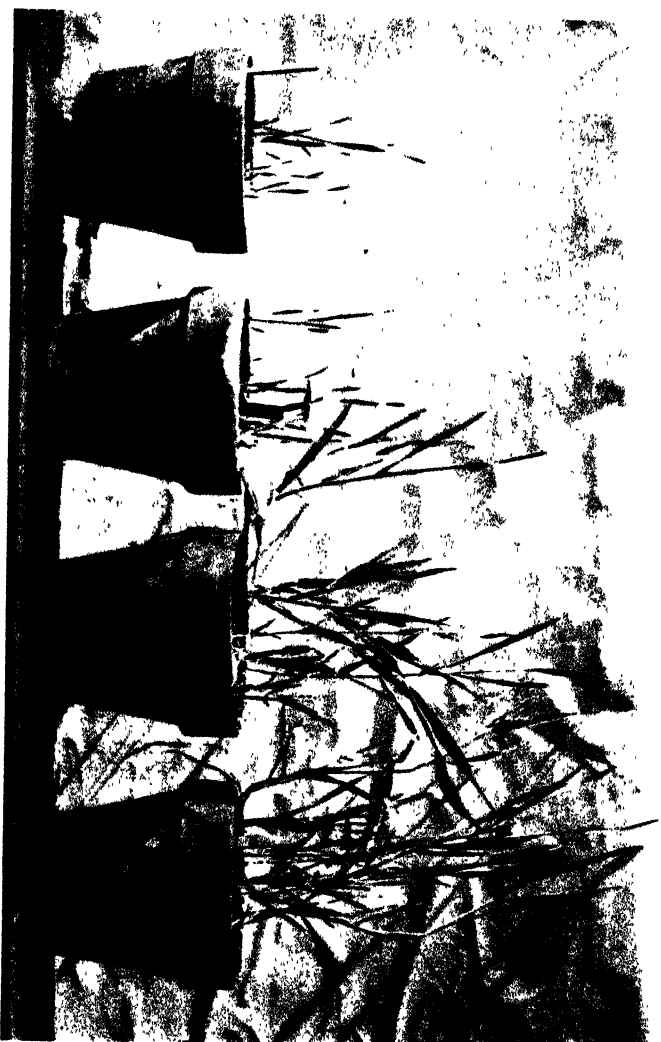


PLATE 4

Treatment of Pots reading from left to right

Pots 16 and 17. 12.00 grams each of  $\text{Na}_2\text{SO}_4$  anhydrous.

Pots 18 and 19. 300.00 grams each of air-dry barnyard manure.



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April 29, 1916

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FUNDAMENTAL INTERRELATIONSHIPS  
BETWEEN CERTAIN SOLUBLE  
SALTS AND SOIL COLLOIDS

BY

L. T. SHARP

UNIVERSITY OF CALIFORNIA PRESS  
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FUNDAMENTAL INTERRELATIONSHIPS  
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SALTS AND SOIL COLLOIDS

BY  
L. T. SHARP

INTRODUCTION

While engaged in an extensive investigation of the physiological effects of  $\text{NaCl}$ ,  $\text{Na}_2\text{SO}_4$  and  $\text{Na}_2\text{CO}_3$  on crop plants as grown in the Davis clay loam, in cylinders, under field conditions, the writer observed that the soil to which the salts had been previously applied became so impervious during the course of the experiment as to retard markedly the rate of percolation. So pronounced was this effect that during the winter and early spring months all of the salt-treated soils were continuously covered with standing water. The appearance of this striking modification in the permeability of the soil to water in the salt-treated soils, together with the inferior cultivating qualities exhibited by them, impressed us as evidence of the fact that the salt treatments under the field conditions of the experiment had effected a fundamental change in the physical constitution of the soil. The occurrence and nature of this change and its relation to soil colloids, interior surface, and other properties of soils, form the considerations with which this paper is chiefly concerned.

Just such an effect on the physical condition of the soil as described above had been anticipated as the normal result of adding  $\text{Na}_2\text{CO}_3$  to the soil, for this salt has generally been conceded, by soil experts, to be an active deflocculating agent. But

to find, on the other hand, that a similar state of diffusion existed in the soils to which  $\text{NaCl}$  or  $\text{Na}_2\text{SO}_4$  had been added appeared wholly inexplicable in the light of the prevailing conception of these salts as agents capable of producing flocculation in the soil colloids. However, this apparent contradiction of well-established facts brought out by the field observations becomes more intelligible as the accumulating data define more clearly the conditions necessary to produce the remarkable effects observed. Thus it has been demonstrated, not only in the field but also in the laboratory, that the removal from the soil by water of  $\text{NaCl}$  or  $\text{Na}_2\text{SO}_4$ , together with the water-soluble products of their chemical reaction with the soil constituents, either wholly or in part, is the initial step in creating a condition favorable for the diffusion of the soil colloids and possibly for the formation of new colloidal matter. Therefore the net result of salt application to and subsequent washing of a soil is to render the soil comparatively impervious and to injure seriously its physical condition. The leaching out of added  $\text{Na}_2\text{CO}_3$  from the soil also presents some interesting phenomena, which are discussed below.

Although the alteration in the physical condition of the soil was first observed by the writer as purely incidental to an investigation primarily designed to ascertain the toxicity limits of the common alkali salts for crop plants, yet it has proved, at least in the case at hand, a most perplexing factor in the production of crops. Our experience would lead us to believe that these after-effects of salt treatments, which appear during the course of leaching the salts from the soil, would have some application to the management of alkali lands, and perhaps some significance with respect to fertilizer treatments. However, the literature on these subjects, with a few exceptions, seems quite devoid of any pertinent reference to the possible importance in these problems of the factor discussed above.

A survey of the literature concerning soils reveals the chief exceptions just mentioned in the following important contributions to this subject. Thus, a brief but significant chapter, "Veränderung der Durchlässigkeit durch Auswaschen der Salze," by Adolph Mayer,<sup>1</sup> records a somewhat sudden reduction

<sup>1</sup> *Forschungen auf dem Gebiete der Agrikultur-Physik*, vol. 2, 1879, p. 251.

in the permeability of a soil when NaOH is washed from it. A similar experiment performed by Mayer with sodium phosphate was not accompanied with the sudden reduction in permeability of the soil as was noted when NaOH was washed from the soil. Likewise the washing out of lime-water produced no marked effect on the rate of percolation, but the leaching out of added NaCl, either with pure water or with lime-water, reduced percolation to a minimum. Mayer ascribes the poor physical condition of drained sea-shore lands to the washing out of the salt, and significantly remarks that this effect, which frequently appears in the second year, is probably more injurious to crop plants than the toxicity of the salt itself.

Van Bemmelen<sup>2</sup> in his classical researches on colloids has also observed a similar decrease in the rate of percolation when loosely bound salts are washed from clays or from the hydrated oxides of tin, silica, and manganese. Moreover, he noted that these colloids, when subjected to salt treatments followed by leaching with water, invariably exhibited a high degree of diffusion upon suspension in water for a second time. He further asserts that this process can be indefinitely repeated by alternately adding to and washing salt from the colloids. The colloidal particles, as remarked by Van Bemmelen, become so exceedingly small, during the process of washing salts from them, as to pass through the filter paper.

Warington<sup>3</sup> also refers in a general way to the appearance of somewhat similar phenomena when soils, previously treated with acids, are washed with water.

It would appear, with the above exceptions, that those who have studied absorption, adsorption, or other physico-chemical effects of salts on soils, have failed to recognize the existence of any relation between the washing out of salts and the subsequent condition of the soil. In fact, Hall and Morison<sup>4</sup> assert that the flocculating effect of salt solutions on kaolin are reversible, that is to say, upon the removal of the added salt the kaolin resumes the original condition of diffusion.

<sup>2</sup> Journ. prakt. Chem., 2nd ser., vol. 23, p. 388, 1881.

<sup>3</sup> Physical Properties of Soil (Cambridge Univ. Press, 1900), p. 30.

<sup>4</sup> Journ. Agri. Sci., vol. 2, p. 244, 1907.

That these earlier findings have a greater significance in agricultural practice than was formerly attributed to them becomes very evident when the practical phase is attempted of cropping the diffused, salt-treated, water-washed soils in our cylinders. Heretofore, the residual effects left upon clays and clay soils after salts have been washed from them have been chiefly considered in connection with their application to the ceramic industry, and have been largely omitted from consideration by those dealing with soils.

Since the striking soil behavior under discussion was first observed by us, as above explained, in crop cylinder experiments, it appears desirable to introduce here a brief description of these experiments.

#### THE CYLINDER EXPERIMENT ON THE PHYSIOLOGICAL EFFECTS OF NaCl, Na<sub>2</sub>SO<sub>4</sub> AND Na<sub>2</sub>CO<sub>3</sub> ON CROP PLANTS

The cylinders used in this experiment are of galvanized iron, open at both ends, coated inside and out with asphaltum, and are fourteen inches in diameter and five feet long. They were placed, during September, 1913, in a clay loam soil at Davis, California, by digging holes to the depth of five feet. During the progress of digging the holes the soil from each foot was carefully removed and set aside separately, thus permitting the cylinders to be refilled with the soil layers in the same order as they exist naturally.

The soils within the cylinders were then supplied with varying percentages of the three sodium salts, NaCl, Na<sub>2</sub>SO<sub>4</sub>, and Na<sub>2</sub>CO<sub>3</sub>. It seemed a difficult task to secure a satisfactory mixture of the salts with the soil already in the cylinders, and it was therefore deemed wise to apply the salts to the surface of the soils by means of a solution containing the weighed quantity of salt in 8800 cc. of distilled water. Accordingly, each cylinder received the same quantity of water, but of a different salt concentration. In addition to the salts some of the soils also received other treatments which, together with the salt treatments, are detailed in the following chart. The quantities of salts were





1913-14 and 1914-15 were accompanied with similar manifestations of the extreme retentiveness and imperviousness of the salt-treated soils.

As previously remarked, all of the check soils responded readily to cultivation and excellent seed beds were easily prepared, while the soils to which  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$  had been added were in an unworkable condition. A crust, an inch or so in thickness, formed as soon as the surface dried, and was so hard that even heavy tools made but little impression on it. Just below the crust the soil was far too wet for plant roots and resembled putty in consistency. Contrary to expectations, the soils treated with  $\text{Na}_2\text{CO}_3$  exhibited better cultivating qualities than the soils receiving the other salts, but inferior to those of the control soils.

With a view to obtaining further information concerning the great dissimilarity between the control soils and the salt-treated soils, it seemed wise to examine some of the surface water standing in the cylinders as well as the soil in contact therewith. These proved to contain but mere traces of any salts. Of next importance a study of the vertical distribution of the salts in the cylinders disclosed the fact that the first few inches of soil contained relatively small quantities of the salts, the first foot more, the second still more, and the third foot most salt. From these results it was concluded that the addition of  $\text{NaCl}$  or  $\text{Na}_2\text{SO}_4$  to a soil and subsequently washing the added salt, or at least part of it, from the soil with water, produces a high degree of diffusion in the soil colloids. Moreover, this alteration in diffusion was accompanied with proportional changes in the other physical characteristics of the soil. These conclusions based on our field experience were soon verified by laboratory experiments.

#### LABORATORY EXPERIMENTS REPRODUCING FIELD OBSERVATIONS

It has been found that the field conditions can be readily reproduced in the laboratory. Thus a simple procedure, illustrating qualitatively the lessened percolation, is to place 10 grams or more of soil in a filter paper in a funnel, adding thereto either  $\text{NaCl}$  or  $\text{Na}_2\text{SO}_4$  as a solid or in a water solution, and then wash-

ing the soil practically free of salt, meanwhile washing with pure water a similar portion of soil from which the salt has been omitted to serve as a control. The difference in the rate of percolation of the two soils has proved to be sufficiently great to be easily discernible even in the case of sandy soils. Frequently

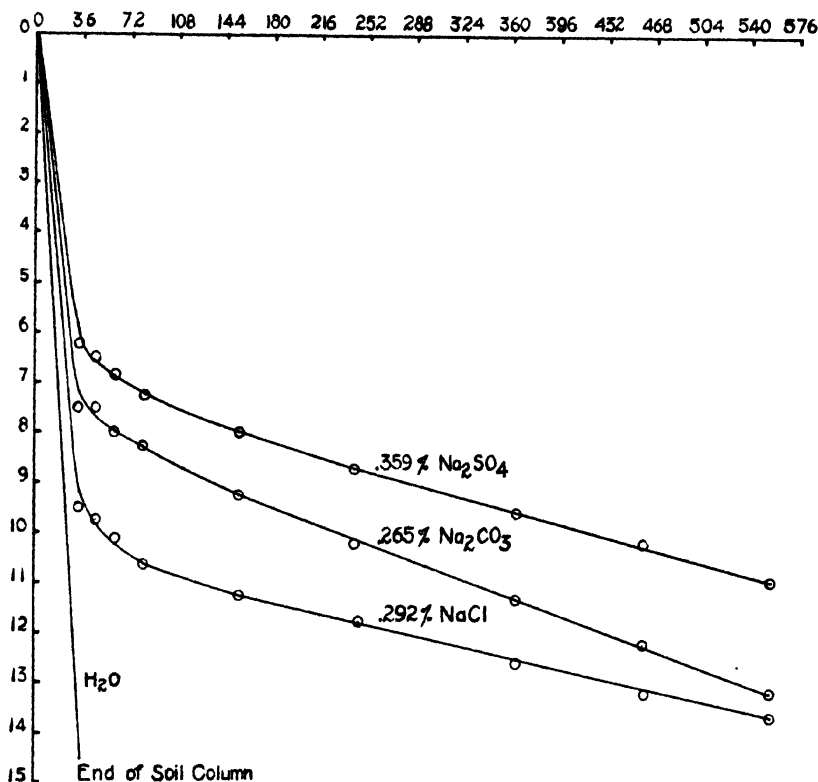


Fig. 2. Curves Representing the Downward Movement of Water Through the Davis Soil to which Salts Have Been Added.

The penetration of the water downward in the soil column is expressed in inches by the ordinates, while the abscissas represent the time of observations in hours.

the fine material mentioned by Van Bemmelen and Warington appears in the percolate.

It has also been found possible to simulate fairly the conditions existing in the field cylinders by means of glass tubes care-

fully filled with the Davis clay loam to which chemically equivalent salt solutions were added, much in the same manner as the salt applications were made to the cylinder soils. A constant head of water was then carefully maintained in the tubes. The downward movement of the water was observed from time to time and is graphically portrayed in Figure 2.

From the curves in it it is very apparent that the influence of these salt additions, accompanied by subsequent surface applications of water to the soil, has been to retard, markedly, the downward movement of the water through the soil. In less than 33 hours the water had penetrated the entire 14½ inches of soil in the control tube, while at the end of 552 hours, under a constant head of three inches, the maximum distance reached in any of the salt-treated soils was 13¾ inches in the case of the soil receiving NaCl, followed by 13¼ inches in the Na<sub>2</sub>CO<sub>3</sub> soil, and but 11 inches in the soil to which Na<sub>2</sub>SO<sub>4</sub> had been added. At the expiration of 56 days no percolation had occurred, though the entire soil columns were moist. The total absence of percolation in the salt-treated soils was attributed to the growth of algae in the tubes.

To test further the points under consideration, a second series was prepared to study the rates of percolation through Davis clay loam to which salts had been added in various ways. The results of this experiment are embodied in Table I.

In the above experiment the chemically equivalent quantities of salts were mixed uniformly with the dry soil before placing the mixture in the paraffine-coated brass percolation tubes, except in the case of Nos. 12, 13, and 14, through which salt solutions were passed of a strength (calculated on the basis of a previously determined water-holding power of the soil) to give comparatively equal quantities of salts with those soils receiving the dry salts. A head of one to two inches was carefully maintained on the soil in all the tubes throughout the experiment.

The results recorded in the foregoing table fully corroborate the field experience. Thus, the average rate of percolation of the check soil was 1.59 cc. per hour, while that of the soil to which NaCl has been added was reduced to .19 cc. per hour. The percolation through the other two soils which received solid

TABLE I

RATES OF PERCOLATION THROUGH SOILS RECEIVING VARIOUS SALT TREATMENTS

Period of percolation ending						Rate of percolation per hour in cubic centimeters											Average of last six Observations		Average of 2nd, 3d, and 4th Observations		Average of 5th, 6th, and 7th Observations	
Period of percolation in hours						This column omitted from all averages																
Period of percolation in hours						This column omitted from all averages																
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20			
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20			
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20			
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20			
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20			
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20			
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1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20			
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20			
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20			
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19</				

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\* At this point an immediate retardation of percolation is noted, due to the substitution of distilled water for the salt solution. The figure 1.66 for No. 12 on Oct. 30, is slightly low due to an oversight in the measurement of the percolate.

$\text{Na}_2\text{SO}_4$  and  $\text{Na}_2\text{CO}_3$  was also decidedly reduced, as shown by the respective percolation rates of .31 cc. and .37 cc. per hour. In significant contrast thereto, the maintenance of a comparatively uniform salt content in the soil column, produced by substituting solutions of the salts for the distilled water as in the case of Nos. 12, 13, and 14, creates a favorable condition for percolation, except in No. 14, which received  $\text{Na}_2\text{CO}_3$ . In this case percolation averaged a rate of 1.07 cc. per hour, which was less than the control, but three times that of the soil receiving  $\text{Na}_2\text{CO}_3$  and distilled water. It is of interest to note the immediate depression in the rate of percolation when distilled water is used instead of the salt solutions of Nos. 12, 13, and 14. Eventually the rate of percolation from these soils under applications of distilled water approached that of the soils originally treated with salts and which in addition had received only distilled water throughout the experiment.

Another percolation experiment, arranged somewhat similarly to the preceding, demonstrated that the relative position of the salt in the soil column, or the manner of adding it, had little or no influence on the depressing effect noted on percolation. It was further observed that a soil, the percolation rate of which had been diminished through the agency of  $\text{NaCl}$  and  $\text{H}_2\text{O}$  applications, failed to recover its original rate of percolation even when a solution of that salt was applied for the second time.

The general trend of the results thus far secured is in accord with Beeson's observations,<sup>5</sup> in which he recorded a delayed absorption of water by soils containing sodium salts as well as a pronounced retarding of percolation through soils to which various salts had been added. Like many other investigators who have observed similar effects resulting from salt treatments, Beeson failed to recognize any connection between the physical condition of the soil and the removal of the salt, but attributed the peculiar changes observed in the soil to surface-tension phenomena, or other alterations in the physical properties of the liquid phase, or, as some investigators are inclined to believe, to a shifting of the soil particles to new positions through the influence of the added salt. Undoubtedly a movement of soil

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<sup>5</sup> Journ. Am. Chem. Soc., vol. 29, p. 620, 1897.

particles does occur when salts are added to soils, but under the conditions obtaining in many of the experiments described in the literature it would seem highly improbable that enough of the added salt remains in contact with the soil to bring about such a movement of the particles. In our experience, the removal of the salt creates among the soil particles a new adjustment which seems of greater importance than that effected by the addition of the salt. Furthermore, if our laboratory experience is properly applicable to field conditions, it would seem of greater advantage to apply  $\text{CaCO}_3$  rather than  $\text{CaSO}_4$  when draining soils containing  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$ , the reverse being apparently true in case of soils impregnated with  $\text{Na}_2\text{CO}_3$ .

#### LABORATORY INVESTIGATIONS OF THE CAUSES OF SALT EFFECTS ON SOILS

The laboratory studies herein reported have been purposely designed to throw some light on the possible causes contributing to the above-noted effects of salts on soils, or at least to ascertain if a relationship exists between the formation of these peculiar physical conditions and the simultaneous occurrence of certain other events. Certain well-known theoretical considerations, reinforced by concrete laboratory experience, have directed the attempts to locate these fundamental causes into three well-defined channels. The first of these is based on an assumption that the salt-and-water treatments have actually increased the quantity of the colloidal matter of the soil. It involves necessarily a study of the soil itself and of the amount and degree of diffusion of the colloidal matter therein contained. The second line of reasoning connects the increase of calcium and magnesium in the percolate from the salt-treated soils, and the absorption of sodium, with the appearance of conditions suitable for the formation of new colloidal matter, as well as with favoring the extreme diffusion of that already present. The third consideration ascribes the diffused condition of the soil colloids to the presence of a small quantity of  $\text{OH}$ -ions in the soil solution. It requires the theoretical assumption that these ions are associated in some manner with the absorption of sodium. Although these

three lines of argument, as outlined, may be closely allied or even parallel, yet in view of our work they seem sufficiently well defined to warrant a separate discussion of each.

At the outset it may be said that it has been extremely difficult to find appropriate and reliable methods of attacking this problem. Certainly opportunity is not lacking for future investigations to perfect accurate methods for definitely measuring certain soil properties or soil constants, which are discussed below. The Davis clay-loam soil was used throughout the following experiments, except as otherwise noted. The designation  $\text{NaCl} + \text{H}_2\text{O}$  used before the word *soil* signifies that the soil was treated with from 0.2 to 0.5 per cent of  $\text{NaCl}$ , most or all of which, together with the soluble salts formed, has been subsequently leached from the soil with water. It must be added that a quantity of Davis clay-loam soil was given the same amount of washing with water to serve as a control soil, and hereafter will be referred to as  $\text{H}_2\text{O}$  soil. A comparison of the  $\text{H}_2\text{O}$  soil with unwashed Davis soil disclosed no important differences.

#### SOME PHYSICO-CHEMICAL OBSERVATIONS ON THE SALT-TREATED, WATER-WASHED SOIL

It seemed reasonable to expect that a study of the  $\text{NaCl} + \text{H}_2\text{O}$  soil itself would reflect, in some degree, the causes contributing to the pronounced imperviousness of such soils. Therefore, as an introduction to this subject, determinations were made of the suspended matter derived from soils which have been subjected to various treatments, and they appear in Table II. The soils and suspending media were placed together in tall hydrometer jars, and were thoroughly shaken for 45 minutes. After standing undisturbed for eighteen hours, aliquot portions of the suspensions were removed by means of a pipette, evaporated to dryness in platinum dishes, dried at  $110^\circ \text{C}$ , and weighed.

The data reported in Table II confirm by an entirely different procedure the original belief with respect to the intensity of the diffusion of the colloids of the salt-treated, water-washed soils. Under the conditions of the experiment 40 grams of normal soil yielded 0.310 grams of suspended matter, while 40 grams of



TABLE II

SUSPENDED MATTER IN SOILS WHICH HAVE RECEIVED VARIOUS TREATMENTS

No.	Davis Soil	Previous Soil Treatment	Suspending Medium	Weight of Suspended Matter-grs.	Per cent of Suspended Matter in soil
1	40 grs.	Washed with distilled water	H <sub>2</sub> O	.3100	.77
2	40 grs.	Boiled in H <sub>2</sub> O	H <sub>2</sub> O	2.5075	6.27
3	40 grs.	.290 grs. of NaCl washed out	H <sub>2</sub> O	2.9555	7.39
4	40 grs.	.....	N/50 Na <sub>2</sub> CO <sub>3</sub>	.2665	.66
5	40 grs.	.....	N/50 NaOH	2.7975	6.99

NaCl + H<sub>2</sub>O soil yielded 2.9555 grams, or nearly ten times the amount of suspended matter found in the untreated soil. It is an interesting coincidence that the rate of percolation previously shown for the untreated soil is almost ten times that for soil to which NaCl has been added and subsequently leached out. It may be properly inferred from this that percolation varies inversely as the degree of diffusion, though our present knowledge does not indicate a relation capable of expression in simple mathematical terms. Furthermore, the inadequacies of the method employed to secure the data in Table II make it impossible to express a positive view with reference to the quantity of colloids in the soils tested, but it is evident that the colloidal matter present is in a much higher state of diffusion in certain of the soils than in the control H<sub>2</sub>O soil. The three treatments, boiling the soil in water, suspending it in NaOH of certain concentrations, and leaching added NaCl from it, produce approximately the same degree of diffusion in the soil colloids, as indicated by the similarity in the results of the quantitative estimations of the suspended matter derived from soils so treated. This agreement in the behavior of the soils receiving the different treatments suggests a similarity or possible relationship between the processes by which these treatments affect the soil or soil colloids. Boiling the soil in water has been assumed by soil physicists to disintegrate the colloidal aggregates. If this be the case and no new colloidal substances are formed by this procedure, then the similarity in colloidal content of the boiled soil and the salt-treated, water-washed soil militates against the sup-

position that the latter treatment has increased the colloidal matter of the soil. On the other hand, boiling the soil in water may bring about a more profound change in the physical condition of the soil than was formerly attributed to it by soil physicists. Some data withheld from publication at this time indicate that the effect of the boiling treatment is of a different nature from that of the salt-and-water treatment, though the soils receiving the two different treatments yield about the same quantity of colloidal matter. On the other hand, the similarity of the colloidal contents of the boiled soil and the soil suspended in NaOH admits of a more plausible explanation on the basis of NaOH as a deflocculating agent.

It was thought that the quantitative data reported above might show sufficient dissimilarity to indicate an actual increase in the soil colloids, but a second series of determinations, reported in a later paragraph, are somewhat contradictory to the above, in that the boiled soil yields a suspension slightly richer in colloidal matter than that derived from a  $\text{NaCl} + \text{H}_2\text{O}$  soil. This point, however, deserves more investigation before a final conclusion is reached.

In the light of certain theories more properly discussed in connection with the third hypothesis, it is of great interest to note the general similarity between the  $\text{NaCl} + \text{H}_2\text{O}$  soil when suspended in  $\text{H}_2\text{O}$  and the normal soil when suspended in NaOH. One might infer that this peculiar agreement in the behavior of the soils in response to two widely different treatments is not accidental. It is also to be observed further that the data under consideration indicates that NaOH and  $\text{Na}_2\text{CO}_3$  are not productive of like results on soil suspensions.

The failure of the suspension method to secure trustworthy results on the quantity of colloidal matter present prompted the adoption of other means for this purpose. But thus far the determination of the hygroscopic coefficient and the dye-adsorption capacity have given negative results, in that they have not indicated any increase in the total interior surface of the soils which have been subjected to the salt treatments. From a theoretical consideration, a soil rich in colloids, or containing colloids in a high state of diffusion, should expose more interior

surface for the deposition of hygroscopic moisture and for dye adsorption, and since these phenomena are presumably direct functions of surface the soils under observation should therefore register increased hygroscopicity and dye adsorption. The findings to the contrary cast some doubt on the validity of these measurements as a reflection of the quantity of colloidal matter present or its degree of diffusion. A third method suggests itself, that developed by Mitscherlich,<sup>6</sup> to study the interior surface through energy exchanges when the soil is moistened with water, but this has not as yet been tried.

The use of the centrifugal machine as employed by Briggs and McLane<sup>7</sup> to ascertain the moisture equivalent of soils in the study of the salt-treated, water-washed soils, has yielded some highly satisfactory results which will be reported in a future paper. The method proposed by Lynde and Dupré<sup>8</sup> for estimating the capillary powers of soils has not proven entirely satisfactory in our hands, when employed for investigating the properties of the salt-treated soils.

It was further questioned whether the physical condition of the soil had been permanently changed or whether the injured soil would completely recover its original condition in response to a second addition of NaCl. Such a supposition naturally implies that some of the reactions involved in producing the increased diffusion partake of the nature of reversible reactions. In order to test this point from a chemical standpoint, it would be necessary to treat the injured soil with its own percolate. Work of this character is reported under the second series of experiments. In this connection, however, the effects of the added salts were considered chiefly in their physical aspects, and accordingly the following experiment was performed: Three grams of NaCl + H<sub>2</sub>O soil were suspended in 10 cc. of NaCl solutions of various concentrations in test-tubes. The time required to clear the supernatant liquid denoted the effect of the NaCl. A similar comparative series with H<sub>2</sub>O soil was also prepared. The results are given in Table III.

<sup>6</sup> *Bodenkunde für Land- und Forstwirte*, p. 51.

<sup>7</sup> U. S. Dept. Agr., Bur. of Soils, Bull. 45.

<sup>8</sup> *Journ. Amer. Soc. Agron.*, vol. 5, no. 2, p. 107, 1913.

TABLE III

EFFECT OF SECOND ADDITION OF NaCl ON TIME REQUIRED TO CLEAR  
SUSPENSIONS OF NaCl+ H<sub>2</sub>O SOIL AND H<sub>2</sub>O SOIL.

No.	Soil	Previous Treatment	Concentration of NaCl as Suspending Medium	Time Required to Clear Suspension
1	3 grs.	NaCl washed out	.....	Almost clear after 600 hrs.
2	3 grs.	NaCl washed out	N/1000	Almost clear after 600 hrs.
3	3 grs.	NaCl washed out	N/500	Almost clear after 600 hrs.
4	3 grs.	NaCl washed out	N/250	Clear after 600 hrs.
5	3 grs.	NaCl washed out	N/100	Clear after 360 hrs.
6	3 grs.	NaCl washed out	N/50	Clear after 20 hrs.
7	3 grs.	H <sub>2</sub> O washed	.....	Clear after 164 hrs.
8	3 grs.	H <sub>2</sub> O washed	N/1000	Clear after 140 hrs.
9	3 grs.	H <sub>2</sub> O washed	N/500	Clear after 117 hrs.
10	3 grs.	H <sub>2</sub> O washed	N/250	Clear after 53 hrs.
11	3 grs.	H <sub>2</sub> O washed	N/100	Clear after 1 hr.
12	3 grs.	H <sub>2</sub> O washed	N/50	Clear after ½ hr.
13	3 grs.	.....	.....	Clear after 140 hrs.

The results of this experiment seemed of sufficient interest to warrant a repetition of the work on a larger scale. Accordingly the following experiment was prepared, much in the same manner as the above, but using 25 grams of soil, 250 cc. of solution, and 250 cc. graduates, instead of test tubes. In this case the weight of the suspended matter was also determined after the mixtures had stood undisturbed for three days. A photograph of this series taken two days after the final shaking is shown (Figure 3).

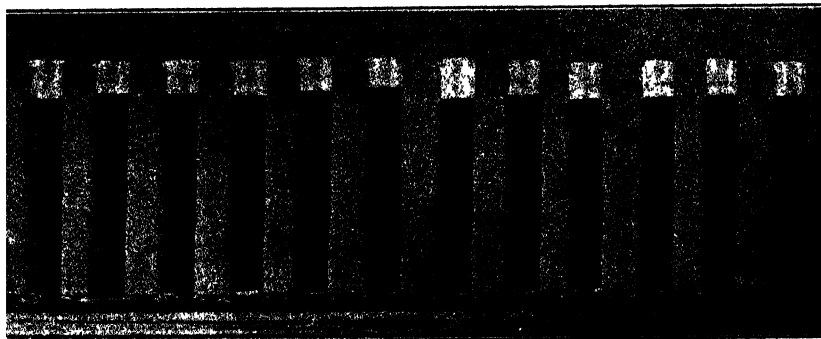


Fig. 3. A Photograph of the Suspensions Described in Table IV

The graduates containing the suspensions as shown in Figure 3 correspond, from left to right, to the Nos. 1 to 12, inclusive, of Table IV.

TABLE IV

EFFECT OF SECOND ADDITION OF NaCl ON THE AMOUNTS OF SUSPENDED MATTER  
FROM NaCl + H<sub>2</sub>O SOIL AND H<sub>2</sub>O SOIL

No.	Soil	Previous Treatment	Concentration of NaCl Suspending Solution	Cc. of Suspending Medium	Weight of Suspended Matter	Per cent of Suspended Matter on Dry Soil
1	25 grs.	NaCl washed out	.....	250	.8345 grs.	3.33
2	25 grs.	NaCl washed out	N/1000	250	.6375	2.55
3	25 grs.	NaCl washed out	N/500	250	.5615	2.36
4	25 grs.	NaCl washed out	N/250	250	.4570	1.82
5	25 grs.	NaCl washed out	N/100	250	.2305	.92
6	25 grs.	NaCl washed out	N/50	250	.0170	.07
7	25 grs.	H <sub>2</sub> O washed	.....	250	.0695	.27
8	25 grs.	H <sub>2</sub> O washed	N/1000	250	.0740	.29
9	25 grs.	H <sub>2</sub> O washed	N/500	250	.0660	.26
10	25 grs.	H <sub>2</sub> O washed	N/250	250	.0615	.24
11	25 grs.	H <sub>2</sub> O washed	N/100	250	Lost, but almost clear	.....
12	25 grs.	H <sub>2</sub> O washed	N/50	250	.....	.....
13	25 grs.	Boiled	.....	250	.9060	3.62

The evidence presented in the last two tables supports the view that the NaCl + H<sub>2</sub>O soil has suffered some physical alteration which is not readily reversed by the second addition of NaCl. The NaCl added to the soil already diffused by previous treatment with that salt and water is here considered as a physical agent, possessing the power to flocculate clay colloids. To test the reversibility of the chemical reactions occurring when salt solutions are allowed to act upon soils requires a soil treatment involving the application of solutions containing, in appropriate form and quantity, the elements removed by the salt applications. Thus, to restore normal conditions in a soil which has been diffused by a salt-and-water treatment would require the replacement of the absorbed sodium by such metals as were originally present in the soil. The larger amounts of NaCl required to flocculate the colloids of the diffused soil may be due to the increased quantity of colloids present, or to the degree of

diffusion of the colloids, or to a change in the nature of the colloid. However, the fact is patent that more NaCl, or a greater length of time for equal quantities of NaCl, is required to produce effects on the NaCl + H<sub>2</sub>O soil commensurate with those on the check H<sub>2</sub>O soil. Table III shows clearly the relative effectiveness of varying concentrations of NaCl in flocculating the colloidal matter as measured by the time required for clearing the suspension. Thus N/100 NaCl flocculates the colloidal matter of the washed soil in one hour, while 360 hours were required to accomplish the same result with the NaCl + H<sub>2</sub>O soil. Nevertheless, the colloidal matter of the NaCl + H<sub>2</sub>O soil seems to be more readily flocculated per unit of NaCl than does that of the H<sub>2</sub>O soil, as is shown in Table IV. Thus the NaCl + H<sub>2</sub>O soil, when suspended in distilled water, yields a suspension containing 0.8345 grs. of solid matter, while the same soil suspended in N/250 NaCl yields but 0.4570 grs. of solid matter. The deposition of 0.3775 grs. of solid matter, in this case, was brought about by 0.058 grs. of NaCl, or at the rate of 6.5 mgs. of solid matter to 1 mg. of NaCl. Similar calculations for the same suspensions of H<sub>2</sub>O soil showed that but 0.14 mgs. of suspended matter was flocculated per mg. of NaCl. Although the NaCl is relatively more effective on the suspended matter of the NaCl + H<sub>2</sub>O soil than on that of the control soil, yet, in but one instance, that of the comparatively strong solution of N/50 NaCl, is the influence of the added salt sufficient to flocculate completely the colloidal matter of the NaCl + H<sub>2</sub>O soil.

Furthermore, it seemed possible that the transformations manifestly occurring in the physical condition of the soil might also be reflected in some measure in the chemical composition of the variously treated soils and of their colloidal substances. Accordingly these materials were subjected to analysis by the strong hydrochloric-acid digestion method, as recommended by Hilgard for chemical soil studies. However, the results secured up to the present time have not confirmed the above presumption. But to what extent future analytical work will enable us to decipher the relationship of the various factors involved in producing the condition under consideration, is still an open question.

TABLE V

RELATION OF CALCIUM AND MAGNESIUM IN PERCOLATE TO SUBSEQUENT COLLOIDAL DIFFUSION

No.	Soil	Amount of Soil	NaCl	Washed with cc. H <sub>2</sub> O	Calcium in Total Washings	Magnesium in Total Washings	Test-Tube Suspensions			
							Soil	cc. H <sub>2</sub> O	Appearance at end of	
						Lost	2 gts.	10	3 days	10 days
1	Anaheim	50 gts.	.00 gts.	1000 cc.	.0058 gts.	.0018 gts.	2	10	Clear	Clear
2	Anaheim	50	.725	1000	.0168		2	10	Almost clear	Clear
3	Berkeley	50	.00	1000	.0028	.0017	2	10	Almost clear	Clear
4	Berkeley	50	.725	1000	.0292	.0155	2	10	Very turbid	Very turbid
5	Davis	50	.00	1000	.0032	.0003	2	10	Almost clear	Clear
6	Davis	50	.725	1000	.0389	.0102	2	10	Very turbid	Very turbid
7	Oakley	50	.00	1000	.0046	.0009	2	10	Clear	Clear
8	Oakley	50	.725	1000	.0104	.0027	2	10	Almost clear	Clear
9	Peat*	25	.00	1000	.0218	.0069	2	10	.....	.....
10	Peat	25	.725	1000	.0324	.0154	2	10	.....	.....

\* The particular peat soil worked with was so light and fluffy that suspensions could not be made.

CHEMICAL STUDIES OF THE CALCIUM AND MAGNESIUM CONTENT OF THE  
PERCOLATE AND THE ABSORPTION OF SODIUM

Our attention was first directed to the possibility of the existence of a relationship between the two factors mentioned in the heading above, by the following experiment, the outline and results of which appear in Table V, page 309.

A review of these data discloses the fact that the application of NaCl to a soil increases the calcium and magnesium found in the percolate as compared with the quantities found in the percolate of the normal soil when leached with distilled water. This is in accord with the results of Kullenberg,<sup>9</sup> Van Bemmelen<sup>10</sup> and the work of others, a good list of which is given by Sullivan.<sup>11</sup> To exemplify the above remarks concerning calcium, let us examine the table with respect to that element. The addition of NaCl to the Anaheim sandy loam and Oakley sandy soil has practically tripled and doubled, respectively, the amount of calcium found in the percolates over that of the percolate from the salt-free soil. In the Berkeley adobe and Davis clay-loam soils the NaCl brings about a much more marked increase of calcium in the leachings. It may be remarked also that the results secured with magnesium are quite parallel to those concerning calcium.

In addition to altering the calcium and magnesium contents of the percolate, the salt treatments materially affected the physical condition of the soil, as shown by a marked retardation of the rate of percolation. All of the soils responded alike to the NaCl treatments, in that they became more impervious and their colloidal matter exhibited a higher degree of diffusion when suspended in water, as shown in the second part of Table V. The degree of imperviousness and diffusion varied, however, with the different soils, and also appeared to be roughly proportional to the increase of calcium and magnesium in the leachings from the soils receiving NaCl. Thus the salt-treated Berkeley and Davis soils, showing notably greater increases in calcium and magnesium in the percolates, were also more highly diffused than

<sup>9</sup> *Jahrb. Fortsch. Agri. Chem.*, vol. 8, p. 15, 1865.

<sup>10</sup> *Landw. Vers. Stat.*, vol. 21, p. 135, 1878.

<sup>11</sup> *U. S. Geol. Surv., Bull.* 312, 1907.



the other two soils, which were much less affected in both these particulars. Evidently the extent of decomposition of the salt by the soil is a factor in determining the final physical condition thereof, and would certainly seem to indicate that the changes of the physical condition of the soil resulting from salt treatments are considerably more than a mere shifting of the soil particles to new positions, as some investigators would lead us to believe. Moreover, it appears proper to infer that these after-effects of salt treatments can be more properly referred to the salt as a chemical agent than as a physical agent.

If it be true that the chemical reaction between soils and salt solutions results in a chemically equivalent exchange of bases, as Sullivan<sup>12</sup> states, then it seems proper to assume that the calcium and magnesium in the leachings from soils treated as described above represent approximately the amount of absorbed sodium. At least it would appear that the calcium and magnesium in the solutions, less the quantity normally present in distilled-water extractions, is an index of the absorbed sodium. This statement has received further justification in some solubility studies, the data of which have not been published. Undoubtedly considerable quantities of sodium have been removed from the salt solutions by the soil, which in return has given up calcium and magnesium. The absorbed sodium has become so firmly fixed in the soil that no amount of washing can dissolve it; otherwise the wash water after passing through the soil would be slightly alkaline. On the contrary, the first portions of the wash water coming through appear to be slightly acid, which is in accord with the work of Sullivan,<sup>13</sup> Parker,<sup>14</sup> and others, but if the washing is continued the leachings eventually become neutral or just perceptibly alkaline, as is the case when distilled water is in contact with the normal Davis soil. These remarks, supported with stronger evidence from the rapidly accumulating literature on chemical exchanges between salt solutions and soils or silicates, point to the formation, under the conditions herein reported, of a sodium alumino-silicate compound, or possibly a series of such

<sup>12</sup> *Loc. cit.*, p. 27.

<sup>13</sup> *Loc. cit.*, p. 8.

<sup>14</sup> *Journ. Agric. Research*, vol. 1, no. 3, p. 1, 1913.

compounds, which resemble most natural silicates in possessing a comparatively inert chemical nature, and which suffer but slight decomposition when in contact with water or are scarcely more than appreciably soluble in water. This hypothesis receives further confirmation in the contention of R. Gans<sup>15</sup> that artificial aluminum silicates behave like natural zeolitic silicates. Moreover, Way<sup>16</sup> clearly recognized the formation of such compounds, and Van Bemmelen<sup>17</sup> is inclined to consider the absorbed salts as fixed in loosely bound chemical combination.

It was originally considered that the substitution of sodium for calcium and magnesium in the soil was a potent factor in bringing about the diffusion of the salt-treated, water-washed soils. However, this first conception attributed the possible effect of the chemical exchange to the double decomposition of the organic salts of calcium and magnesium by the NaCl, which would result in the formation of  $\text{CaCl}_2$  and  $\text{MgCl}_2$  and organic compounds of sodium. Since these end-products are all soluble in water, a continuous leaching of the soil with that solvent would be likely to deprive the soil of a portion of its organic matter and of a certain amount of calcium and magnesium. If the organic matter herein referred to can be properly catalogued under those elastic and indefinite terms, humus or humates, then it appears reasonable in the light of Schloesing's work<sup>18</sup> to expect that the physical condition of the soil, depleted of such organic matter, would be seriously affected in the direction already suggested. The loss of such well-known flocculating agents as calcium and magnesium might also be reflected in the changed physical aspect of the soil.

But the cogency of the argument just presented is considerably lessened by the results of the experiment to be described next. Twenty-five gram portions of the Davis soil were subjected to the treatments outlined in Table VI. The soils were then allowed to dry, after which 10 grams of each soil were suspended in 100 cc. of distilled water for 48 hours before the sus-

<sup>15</sup> Cited from *Exp. Sta. Rec.*, vol. 31, no. 1, p. 22, 1914.

<sup>16</sup> *Journ. R. Agric. Soc.*, vol. 13, p. 123, 1852.

<sup>17</sup> *Loc. cit.*; also in *Landw. Vers. Stat.*, vol. 35, p. 121, 1888.

<sup>18</sup> Cited from Hilgard, *Soils* (1911), p. 111.

TABLE VI

THE EFFECT OF REMOVING HUMUS FROM THE SOIL IN ITS RELATION TO THE  
DIFFUSION APPEARING IN THE SOIL AFTER SALT AND  
WATER TREATMENTS

No.	Soil	Treatment	Weight of Suspended Matter from 10 grams in 100 cc. of water
1	25 grs. (Davis soil)	Washed with H <sub>2</sub> O	.0184
2	25 grs. (Davis soil)	50 cc. 4N HCl then washed with H <sub>2</sub> O	.0118
4	25 grs. (Davis soil)	HCl,* H <sub>2</sub> O, NH <sub>4</sub> OH, and H <sub>2</sub> O	.0250
5	25 grs. (Davis soil)	HCl, H <sub>2</sub> O, NaCl, and H <sub>2</sub> O	.1216
6	25 grs. (Davis soil)	HCl, H <sub>2</sub> O, NH <sub>4</sub> OH, H <sub>2</sub> O, NaCl, and H <sub>2</sub> O	.1842
7	25 grs. (Davis soil)	NH <sub>4</sub> OH	.0176

\* The symbols used herein designate the soil treatments on the filter paper with the various substances; as indicated, H<sub>2</sub>O applications were generally made between applications of the other materials. Solutions approximating normal strength were generally employed.

pension above the deposited material was drawn off. An aliquot of this suspension was evaporated to dryness, gently ignited, and weighed to obtain the data of column 4, in Table VI.

The weight of the suspended matter secured from suspensions of Davis soil which had been subjected to the various treatments gives an index of the extent of the physical effects of such treatments. The treatment with HCl, followed by washing until the filtrate was practically free from chlorides, seemed to reduce the quantity of material capable of being held in suspension, yet the soil had no doubt lost a considerable quantity of its original calcium and magnesium content. This fact certainly indicates that the loss of calcium and magnesium from the soil bears but little relation to the diffusion of the soils as a general proposition.

The soil treatment in the case of No. 4, which simulates the procedure generally employed in humus determinations, with the exception that water is used as a final application, gives a suspension slightly richer in solids than the soil treated with water alone. If, in addition to the treatment just mentioned, NaCl is then added to the humus free soil,<sup>19</sup> the suspension becomes a very turbid liquid rich in solids in a highly diffused state,

<sup>19</sup> The term humus here signifies that portion of the soil's organic matter removed by the treatment in vogue for that purpose.

as shown in the case of No. 6. Evidently the loss of humus and its organic colloids increases, rather than decreases, the amount of material finally found in suspension, for No. 5, receiving HCl and NaCl treatment, yields a suspension containing less colloidal material than does No. 6. In all probability the increase of colloidal material in No. 6 over No. 5 is in no way connected with the loss of organic matter of No. 6, but is more likely to be due to the conditions and treatments involved in extracting the humus. Furthermore, the organic colloids of soils seem to have but little direct relation to the phenomena which appear when certain salts are washed from soils.

That the deflocculated condition of the salt-treated soils might be due in some measure to the loss of calcium and magnesium from the soil, in a manner somewhat analogous to that outlined by Foerster,<sup>20</sup> appeared as a secondary consideration. The actual loss of calcium and magnesium, however, cannot be considered the sole factor in producing the striking conditions observed. For, if that were the case, the application of acids followed with washing should bring about similar results. On this point Warrington has already noted that normal diffusion of the colloids will reappear in a soil which has once been flocculated by acids when the acids and such salts as they have formed are removed by washing. Our experience with soils treated in the manner just described fails to show any marked increase in colloids, as is the case when the soil is treated with NaCl. The treatment with HCl followed by leaching with water seems to retard to a notable extent the rate of percolation through the Davis soil. However, if any new colloids are formed by this treatment, or if the original colloidal content of the soil is thoroughly diffused thereby, as the case may be, then these effects are entirely obliterated by the subsequent drying of the soil, for a suspension of the dry soil so treated yields no additional colloidal matter. This fact tends to confirm the view that the loss of calcium and magnesium is not alone responsible for the diffusion of the soils.

Furthermore, the writer has shown clearly by a rather simple procedure that a considerable exchange or a direct addition of bases is essential for the production of the diffused condition.

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<sup>20</sup> Chem. Ind., vol. 28, no. 24, p. 733, 1905.

A solution of N/10 NaCl was allowed to act upon successive portions of Davis soil until practically no more calcium or magnesium was taken up by the solution, so that a solution of chlorides was formed which was quite inert toward this soil. A solution secured in this manner appears to approach the saturation point with respect to calcium and magnesium compounds of the soil. This solution remained practically N/10 with respect to chlorine throughout its successive periods of contact with the soil, and at no time was the solution entirely depleted of its sodium content by the interchange with calcium and magnesium. The application to the Davis soil of a solution secured in the fashion described above and followed by washing with water did not result in such pronounced diffusion as was observed upon the application of N/10 NaCl solution followed by washing with water. It may therefore be inferred that the chemical exchange of bases plays a significant role in the mechanism by which diffusion is produced in soils which have been washed after an addition of NaCl.

By the various lines of reasoning outlined above, the majority of the more obvious possible causes for the extreme diffusion of the soils under discussion have been completely eliminated or at least reduced to factors of little significance. For this reason, more mature thought on the subject has directed our attention to the absorption of sodium, which has been previously mentioned, as the keynote to the deteriorated physical condition of the salt-treated, water-washed soils. The substitution of sodium for the calcium, magnesium, or other bases in the silicate complex, or the direct addition of sodium to such a complex, results, according to our proposed hypothesis, in the formation of new jelly-like colloids, capable of becoming highly diffused when in contact with water. The fact that leaching  $\text{Na}_2\text{CO}_3$ , NaOH,  $\text{NaHCO}_3$ , NaCl,  $\text{Na}_2\text{SO}_4$ , and  $\text{NaNO}_3$  from the soil brings about the same appearance of the soil and the same physical manifestations of deflocculation, becomes more comprehensible if the view is accepted that the absorption of sodium and the new compounds formed thereby are, in the main, the factors responsible for the deflocculated condition of the soils so treated. Moreover, these various treatments are accompanied by the appearance of widely

different quantities of calcium and magnesium in the percolate, thus affording additional evidence that the increased solubility and the consequent loss of these elements from the treated soils is not the prime factor in causing the ultimate diffusion of the soil colloids. The facts herein presented concerning the absorption of sodium and the formation of new colloidal substances corresponds in the main with the idea of unsaturated silicates and absorption phenomena, as developed by Cameron,<sup>21</sup> Harris,<sup>22</sup> and Loew,<sup>23</sup> and others, in their work dealing with acid soils.

The conception of the causes of the apparent deflocculation, as discussed above, receives tangible substantiation in the results of the following experiment. Twenty grams of Davis soil were subjected to the treatments outlined in Table VII. After washing with water, the soils were dried at room temperature and then 10 grams of each were suspended in 100 cc. of water to secure the figures in the last column. Meanwhile the calcium and magnesium in the total percolates were determined in the usual fashion.

TABLE VII  
EFFECTS OF WASHING VARIOUS SODIUM SALTS FROM THE DAVIS SOIL

No.	Soil (Davis)	Treatments	Calcium in Total Percolate grs.	Magnesium in Total Percolate grs.	Weight of Suspended Matter from 10 grams in 100 cc. of water
1	20 grs.	H <sub>2</sub> O only	.0017	.0014	.0348
2	20 grs.	100 cc N/10 NaOH fol- lowed with water	.0015	.0005	.4860
3	20 grs.	100 cc N/10 Na <sub>2</sub> CO <sub>3</sub> fol- lowed with water	.0020	.0037	.6500
4	20 grs.	100 cc. N/10 NaCl fol- lowed with water	.0108	.0109	.4720
5	20 grs.	100 cc. (approx.) N/10 NaHCO <sub>3</sub> followed with water	.0035	.0086	.4880
10	20 grs.	100 cc. N/10 NaNO <sub>3</sub> fol- lowed with water	.0088	.0103	.4340

It is clear that all of the sodium salts used in the above experiment produce approximately the same effects on the colloidal matter of the soil when washed from it, as indicated by the

<sup>21</sup> The Soil Solution (Chemical Publishing Co.), 1911.

<sup>22</sup> Journ. Phys. Chem., vol. 28, no. 4, p. 355; and Michigan Agric. Exper. Sta. Tech. Bull. 19.

<sup>23</sup> Porto Rico Agric. Exper. Sta., Bull. 13, 1913.

amount of material finally found in suspension. All of the salt-and-water treatments yielded suspensions containing more than ten times the suspended matter found in the water-washed soil. From the evidence reported in the above table it seems proper to infer that neither the nature of the added sodium salt, nor the loss of calcium and magnesium from the soil, have much part in the production of the deflocculated condition noted in the salt-treated, water-washed soils. The data of Table VII are also discussed later in their relation to the possible effects of NaOH and the OH-ion on the physical condition of soils.

The interchange of ions between the soil silicates and neutral salt solutions like NaCl or  $\text{Na}_2\text{SO}_4$ , which seems to result in the formation of new colloidal substances, also necessitates the simultaneous presence of free acid or of new salts of the free acid as the calcium or magnesium salts, in the solution bathing the soil particles. As previously discussed, both of these conditions have been encountered in the supernatant liquid of a neutral salt solution in contact with soil and also in the first portion of the percolate coming through a soil to which a neutral salt has been added. The constant presence of the free acid and its soluble salts, together with more or less of the salt originally added, occasions the maintenance of a flocculated condition of the soil colloids, so that any additional colloidal matter which may have been produced is not sufficiently effective on the physical character of the soil to be easily recognized.

If it be assumed that new and additional colloidal material is formed in the soil by virtue of the salt-and-water treatments, then it is most likely in existence prior to the washing process, but its effects on the physical condition of the soil are not manifested until the surrounding medium has become sufficiently dilute, with respect to salts, to allow of a more or less complete diffusion of the soil colloids. The addition of the neutral salts either produces new colloidal matter simultaneously with the chemical interchange of bases, which occurs independently of the washing with water, or in some manner disintegrates the existing colloidal aggregates. The former seems the more plausible, especially in view of certain phases of the work herein presented. In addition we also have evidence that the washing with water alone does not

measureably increase the colloidal content of the soil, nor does it materially affect its physical condition.

At least one interpretation of these facts seems plausible, namely, that the neutral salts or their ions function as the creative agent whereby diffusible colloidal matter is formed, while the washing with water serves in the entirely separate capacity of removing from the sphere of activity any flocculating agents in the shape of soluble salts which may have been present.

Certain modifications of the hypothesis just presented must be considered in order to account for the action on soils of the salts which give an alkaline reaction. First of all, the chemical products that can possibly be formed when the latter class of salts is allowed to act upon soils are on the whole comparatively insoluble and hence possess relatively small flocculating powers. Owing to this fact the washing process, which seems to be essential for the appearance of the diffusion in soils treated with neutral salts, is not such an important factor in case of soils treated with salts giving rise to an alkaline reaction.

Instead of attributing the deflocculation of soils which have received NaOH or alkaline carbonates, wholly to the OH-ion content of the solution or to the alkaline reaction so produced, our present theory, supported by the facts already presented, proposes to account for the diffusion of the soils so treated by the formation of colloidal sodium alumino-silicate complexes under conditions which permit of an immediate deflocculation. Therein lies the difference in the behavior of neutral salts on soils, as compared with that of salts of an alkaline reaction. In all probability somewhat similar compounds are formed in the two cases, but in the first case, with the exception of certain circumstances, the conditions are such as to prevent deflocculation, while in the second case deflocculation is at least permitted and perhaps accentuated.

#### A POSSIBLE RELATION BETWEEN THE COMPOSITION OF THE SOIL SOLUTION AND THE DIFFUSION PHENOMENA IN CERTAIN SALT-TREATED, WATER-WASHED SOILS

The first two hypotheses formulated to explain the diffused condition of the salt-treated soils dealt largely with the soil itself and with the absorption of sodium by the soil. A third hypothesis



proposes to attribute the diffusion of the soil colloids to changes in the composition of the medium, namely the soil solution. Such a diffused condition of the soil colloids might be brought about by an increase in the OH-ion content of the solution bathing the soil particles. This concept is based on the fact that clay is a negatively charged colloid, and according to the views now held the further addition of negative ions to such a colloidal suspension causes these particles to assume greater charges of like sign, so that they repel each other and thus remain distributed throughout the medium in a stabilized condition. On the other hand, it is held that the introduction of ions bearing an opposite charge to that of the colloidal particles neutralizes the charge associated with the particles, so that they no longer repel each other but gather together in aggregates or floccules. For a more complete discussion and bibliography dealing with these phenomena, the reader is referred to the work of Whitney and Ober.<sup>24</sup> In a more recent review Tolman<sup>25</sup> has advanced a clear conception of colloids and their behavior, which affords us a satisfactory working basis for studies on these substances. According to this author, the surface tension existing between the particles and the surrounding liquid is the factor which determines the degree of dispersion of the particles in the liquid. Thus systems of zero surface tension are at equilibrium. Those possessing a negative surface tension increase, automatically, their degree of dispersion until the zero value is reached, while those of positive surface tension tend to become less dispersed. Since the surface tension referred to is the resultant of many forces, it may be readily affected in numerous ways, as by the mechanical process of grinding, by heating, by the addition of electrolytes, or by the passage of an electric current. Through the application of these considerations we may be able to decipher more clearly and definitely the problems involved in the effects of salts on the physical condition of soils.

It is possible that deflocculants other than the OH-ion may have been introduced into the soil solution by means of the salt-and-water treatments, but the latter factor obviously appears as the most significant deflocculating agent likely to be present

<sup>24</sup> Journ. Amer. Chem. Soc., vol. 23, p. 842, 1901.

<sup>25</sup> Journ. Amer. Chem. Soc., vol. 35, no. 4, 1913.

under the conditions of the experiments and hence merits first attention. Probably the most striking evidence of the possibility that the OH-ions in the soil solution may be justly deemed the cause of the diffusion of the soils with which this paper is chiefly concerned, lies in the well-known deflocculating effect of dilute solutions of NaOH on the soil colloids. Thus the poor tilth and cultivating qualities of lands impregnated with black alkali ( $\text{Na}_2\text{CO}_3$ ) has been rather vaguely attributed to the OH-ions derived from the hydrolysis of the  $\text{Na}_2\text{CO}_3$ . This expression fails to offer any explanation of the mechanism whereby the OH-ion induces the observed effects, neither does it allow of any possible effect of the Na-ion on the physical properties of the soil. However, if our interpretations be not too far amiss, there are several reasons, not based on theoretical considerations alone, but substantiated by facts, for suspecting that the OH-ion is of much smaller significance than the accompanying Na or other basic ion in the final effect of the chemical compound on the physical condition of the soil.

The common conception that substances which yield an alkaline reaction on hydrolysis occasion the deflocculation of the soil colloids is frequently accepted without qualification, even by those working with alkali soils, notwithstanding some facts now extant which deny its validity. Thus, as early as 1874 Durham<sup>26</sup> pointed out that clay suspensions cleared more rapidly in strong  $\text{Na}_2\text{CO}_3$  solutions than in distilled water. Whitney and Straw<sup>27</sup> have also shown that NaOH in dilute solutions tends to stabilize suspensions of colloidal silver, china clay, and lampblack, and that emulsions of turpentine, carvene, and carvol are also acted upon in a similar manner. The behavior of these substances also gave evidence that the maximum stability occurred at certain concentrations of NaOH, above which flocculation was produced and below which the effect of the NaOH was not so pronounced. The investigations of Hall and Morison already referred to substantiate, in the main, the previous citations on the point under discussion. Quite recently Maschhaupt<sup>28</sup> has found that even

<sup>26</sup> Chem. News, vol. 30, no. 676, p. 57, 1874.

<sup>27</sup> Journ. Amer. Chem. Soc., vol. 29, p. 325, 1907.

<sup>28</sup> Landw. Vers. Stat., vol. 83, p. 467, 1914.

0.015 N NaOH has a flocculating effect on the colloidal matter of a sandy loam soil, while more dilute solutions of NaOH stabilized the diffusible colloidal matter. He further asserts that alkali carbonates act much in the same manner. Despite the preceding evidence, Rohland<sup>29</sup> still contends that the flocculation of clay soils by lime is due to the OH-ion.

While working along similar lines, the author has noted that a suspension of the Davis clay loam settles out more rapidly in a 0.05 N solution of NaOH than in distilled water, but that solutions of greater dilution were stabilizing in their effects. On the other hand, no solution of  $\text{Na}_2\text{CO}_3$ <sup>30</sup> proved effective as a stabilizing agent as compared with distilled water, while solutions of  $\text{Na}_2\text{CO}_3$  stronger than 0.022 N had a decided flocculating effect. This difference in the behavior of NaOH and  $\text{Na}_2\text{CO}_3$  has not come to the writer's attention before in the literature of the subject, and fails to support the widespread teaching that  $\text{Na}_2\text{CO}_3$  and salts which hydrolyze similarly deflocculate the soil colloids through the agency of the OH-ion. But the most striking feature of the action of NaOH on the soil suspension was the marked resemblance of the soils suspended in that medium to the  $\text{NaCl} + \text{H}_2\text{O}$  soil suspended in distilled water. The yield of suspended matter from these two suspensions proved to be of about the same magnitude, as shown in Table II. A cursory consideration of this fact obviously supports the contention that the OH-ion content, or the alkalinity of the suspending solution, may be responsible for the diffusion of the soil colloids in the cases under consideration. However, an interesting point to the contrary lies in the fact that suspensions of the Davis soil in very dilute solutions of NaOH can not be distinguished from suspensions of the same soil in distilled water. That is to say, NaOH solutions of a concentration of N/1500 to N/2000 exercised no recognizable effect on the soil colloids. The concentrations here referred to approach the same order of alkalinity as that found in the solution bathing the particles of the highly diffused  $\text{NaCl} + \text{H}_2\text{O}$  soil. Evidently some other factor than the OH-ion

<sup>29</sup> Landw. Jahrb., vol. 44, no. 3, p. 437, 1913; and Landw. Vers. Stat., vol. 85, nos. 2, p. 123, 1914.

<sup>30</sup> Baker's analyzed  $\text{Na}_2\text{CO}_3$  was used in these experiments except where otherwise noted.

content of the solution is either responsible for the diffused condition of the soil colloids, or at least is of material assistance in producing this effect.

Moreover, it has been demonstrated that the washing out of the excess NaOH does not materially benefit the injured physical condition of the soil, although the alkalinity of the soil solution was thereby reduced to a concentration comparable with that of the dilute solutions to which reference has just been made. Actual determinations of the alkalinity of the final portions of the percolate from soils to which NaOH has been added show only such alkalinity as the percolate from the normal soil. If we accept the view brought forth by Cameron,<sup>31</sup> which seems justifiable, then the percolates from these soils approach, as a limit, the chemical composition of the soil solution and hence we may conclude that the alkalinity of the soil solution is of the same order of magnitude as that of the percolate. The fact that washing the NaOH from the soil is not accompanied by an improvement in the physical condition of the soil may be fairly interpreted as substantially affirming the view that the OH-ion is of little moment in the diffusion of the NaCl-| H<sub>2</sub>O soil, and possibly has but little connection with the deflocculation of soils to which NaOH has been added. Furthermore, the facts just discussed, taken in conjunction with those considered under the heading of sodium absorption, lend an appearance of reality to the assumption that the sodium, even in the case of direct addition of NaOH to soils, is the principal agent in creating the diffused condition in soils so treated.

Moreover, the writer has found that NaHCO<sub>3</sub>, Na<sub>2</sub>CO<sub>3</sub>, and NaOH have distinctive effects on the soil colloids. In higher concentrations all three salts prove to be flocculants; in very weak concentrations, as of the order of N/2000 or less, they seem to resemble distilled water in their behavior toward the soil colloids. In medium concentrations, that is, those less than 0.05 N, NaOH is a deflocculating agent, Na<sub>2</sub>CO<sub>3</sub> acts much like distilled water, while NaHCO<sub>3</sub> seems to be a pronounced flocculant. At least the NaOH and Na<sub>2</sub>CO<sub>3</sub> yield a certain amount of OH-ions, and in the wide range of concentrations employed some

<sup>31</sup> Eighth Intern. Cong. Appl. Chem., vol. 15-16, p. 49, 1912.

point must have been encountered at which the OH-ion concentration of these two was the same or approximately so. Yet the two salts exercised a different influence on the soil colloids at all the concentrations used, with the exception of the highest and lowest concentrations. This fact indicates that some other factor than the OH-ion is effective in determining the degree of dispersion of the soil colloids. What application this may have to black alkali lands is a question, for under natural conditions it is most likely that all three compounds, NaOH, NaHCO<sub>3</sub>, and Na<sub>2</sub>CO<sub>3</sub>, and their respective ions occur.

But the most striking feature of this phase of the problem lies in the fact that the washing out of the soluble matter from separate portions of the Davis soil receiving NaOH, Na<sub>2</sub>CO<sub>3</sub>, and NaHCO<sub>3</sub> results in the same way. The soil becomes very impervious and diffuses when shaken with distilled water. Thus the cylinder soils receiving Na<sub>2</sub>CO<sub>3</sub> have been exposed to conditions permitting the leaching out of the soluble salts, at least from the surface soil, so that they now exhibit the same peculiarities as the soils treated analogously in the laboratory.

The systems so far considered have been largely made up of soils to which various quantities of NaOH, Na<sub>2</sub>CO<sub>3</sub> or NaHCO<sub>3</sub> have been added. To ascertain to what extent the facts so gained are applicable to the conditions existing in the laboratory samples of NaCl + H<sub>2</sub>O soil or to those of the field cylinder soils receiving NaCl and Na<sub>2</sub>SO<sub>4</sub>, necessitates the measurement of the alkalinity obtaining in these soils. We have attempted to secure some information with regard to the quantity of OH-ions or the alkalinity in the solution containing a suspension of the NaCl + H<sub>2</sub>O soil, but the persistent color of this solution has made it impracticable to employ it directly with the various indicators. It would seem, however, from actual titrations made in the usual manner using methyl orange as the indicator, that the alkalinity of the soils in the cylinders receiving NaCl and Na<sub>2</sub>SO<sub>4</sub> had been somewhat increased over that of the control soils. But solutions secured from soils treated in the laboratory in a manner similar to the treatment of the field soils failed to verify consistently the above observations. At the present time we are engaged in a study of the reaction of the soil suspensions by the use

of the hydrogen electrode. The results thus far secured indicate that there is not a sufficient quantity of OH-ions in the NaCl + H<sub>2</sub>O soil to produce the degree of deflocculation observed therein.

If the OH-ion content of the medium is assumed to be the causal agent for the diffusion of the soil colloids, it would appear an equally plausible assumption that the transference of the suspending medium from a diffused soil to the original untreated soil would carry with it the deflocculating agent, so that the second soil would register to some degree the physical manifestations of deflocculation. To test the correctness of this hypothesis the following experiment was undertaken. Twenty-five grams of NaCl + H<sub>2</sub>O soil were suspended in 250 cc. of water. The colloidal matter of such a suspension normally remains in a stabilized condition for at least three or more weeks, when exposed to laboratory conditions. This suspension was cleared, however, at the expiration of 48 hours by passing it through the Pasteur Chamberland filtering candle under pressure. This was accomplished with considerable care so as to conform to the conditions found by Briggs<sup>32</sup> to yield the most satisfactory results. The solution secured in the manner described from the NaCl + H<sub>2</sub>O soil was then used as a suspending medium for 10 grams of untreated Davis soil. A comparison of the suspension thus formed with a suspension of the same soil in distilled water disclosed no recognizable difference. Thus it would appear that a transference of the suspending medium of a diffused NaCl + H<sub>2</sub>O soil to a second untreated soil does not carry with it the agent causing the deflocculation of the first soil. Hence, any hypothesis ascribing the altered physical condition of the NaCl + H<sub>2</sub>O soil to changes in the soil solution seems untenable.

The value of the results secured by the procedure outlined above may be open to question. To avoid such criticism, the writer made the following experiment, which distinctly shows that the transference of a soluble soil deflocculant from one soil to another by the method employed is susceptible of proof. A N/50 NaOH solution in contact with the Davis soil tends to maintain the soil colloids in a stabilized condition. Such a diffused suspension was subjected to the filtering treatment out-

<sup>32</sup> U. S. Dept. Agr., Bur. of Soils, Bull. 19, p. 31, 1902.

lined above and on passing through the Pasteur filter yielded a clarified solution which had not lost its power to deflocculate the Davis soil. The writer is fully aware that conditions might arise wherein such treatments would considerably modify the deflocculating power of the solute—for example, in case of very dilute solutions where absorption by the soil and filter would be relatively large and possibly of sufficient magnitude to markedly diminish the quantity of deflocculant in the filtrate, which would result undoubtedly in a decrease in the deflocculating power of the solution. A similar reduction in deflocculating power of a solution would also be likely to appear if a solution were subjected to repetitions of the procedure described above.

Despite the evidence above some doubt may still be entertained as to the absence of significant quantities of OH-ions in the films of water on the immediate surfaces of the colloidal particles, especially in view of the probability that the colloidal matter in the diffused soils consists largely of compounds of the chemical nature of sodium silicate, which hydrolyzes to some extent in water and eventually gives rise to OH-ions, thereby lowering the surface tension as described by Tolman<sup>33</sup> and likewise producing the conditions obtaining in the "natant" colloids of Hall and Morison.<sup>34</sup> Moreover, the results secured by Briggs<sup>35</sup> on the absorption of alkali hydrates by silica tends to confirm the proposed conception of "natant" colloids. If this be the case, the rate of diffusion of these ions from the films into the more dilute medium would be the factor determining whether it would be possible to transfer a sufficient quantity of these ions, by means of the solution, to be effective on the physical condition of a second, otherwise untreated, soil. Under the conditions of the experiment just cited, a contact period of two days was allowed for such diffusion, which would appear to allow ample time therefor.

Furthermore, as observed by Whitney and Straw<sup>36</sup> and others, the fact that NaOH in certain concentrations tends to stabilize

<sup>33</sup> *Loc. cit.*

<sup>34</sup> *Loc. cit.*

<sup>35</sup> *Journ. Phys. Chem.*, vol. 9, p. 617, 1905.

<sup>36</sup> *Loc. cit.*

colloidal matter of a comparatively inert chemical nature forms the basis of an argument which opposes the conceptions herein presented, and which favors the view that the OH-ion may, after all, play an important role in the diffusion of colloids. Thus, systems composed of colloidal silver, lampblack, or relatively pure organic substances in contact with NaOH probably do not offer opportunity for the direct addition of the sodium or the exchange of ions whereby sodium is taken up, a factor which forms an essential link in the scheme proposed by the writer to explain the effects of the alkali salts on soils. Also the experiments of Bliss, cited by Whitney and Straw,<sup>37</sup> give an indication that NaOH may be an effective agent, through the medium of the OH-ion. No attempt is made, however, to deny the complete ineffectiveness of the OH-ion, for it undoubtedly has an important influence on the physical condition of colloids, as is the case with many ions. But with regard to the physical condition of the salt-treated Davis soil, it is very evident that the OH-ion is a factor of much less significance than the other ions associated with it.

#### THE PRECIPITATING EFFECT OF VARIOUS ACIDS AND SALTS ON THE SOIL COLLOIDS

There seemed some possibility that the presence of some ion or ions, other than the OH-ion, in the soil solution might be held accountable for the extreme diffusion of the  $\text{NaCl} + \text{H}_2\text{O}$  soil and soils similarly affected by other salt treatments. Consequently an extensive series of test-tube experiments dealing with the effects of various acids and salts on the soil suspension were undertaken. The results of these tests are briefly referred to at this time in their relation to the condition existing in the salt-treated, water-washed soils.

It was found that N/2000 HCl and  $\text{H}_2\text{SO}_4$  perceptibly flocculated the soil colloids, as compared with distilled water suspensions of the same soil. To attribute this action wholly to the H-ion of the acid would, in the writer's opinion, be erroneous, for no doubt salts are immediately formed when the acid comes

<sup>37</sup> *Loc. cit.*



into contact with the soil. Nevertheless, it is significant that acids of the low concentration here employed are more efficient flocculants than their salt solutions of similar concentrations. Hence the unneutralized acid in very dilute concentrations undoubtedly exercises a pronounced flocculating power, so that this factor is eliminated as the possible deflocculant in the case of  $\text{NaCl} + \text{H}_2\text{O}$  soil.

Solutions of the chlorides, sulphates, and bicarbonates of calcium, sodium, ammonium, and potassium, were also studied with regard to their effects on the colloidal matter of soils. These salts in solutions ranging in concentration from N/1500 to N/500 possessed distinct flocculating powers. Solutions of higher concentrations were likewise flocculating in their effect, while more dilute solutions behaved similarly to distilled water. Hence the diffused condition of the  $\text{NaCl} + \text{H}_2\text{O}$  soil cannot be attributed to the mere dilution of the neutral sodium salts or of the simple salts formed by reactions between the added salts and the soil silicates. As previously indicated, this statement must not be construed to include the systems soil +  $\text{NaOH}$ , or soil +  $\text{Na}_2\text{CO}_3$ , but must be considerably modified to express the facts existing in those cases.

#### THE EFFECTS OF WASHING VARIOUS SALTS FROM THE SOIL WITH WATER

The washing out of  $\text{KCl}$  and  $\text{NH}_4\text{Cl}$  from the Davis soil with water gave essentially the same results as accompanied the removal of the sodium salts by the same means. On the other hand, the leaching out of calcium salts seemed to leave the soil colloids in a more flocculated condition than in the normal, untreated soil, though this effect was by no means so pronounced as when the calcium salts were allowed to remain in contact with the soil particles. According to the ideas herein presented, the effect of a salt solution, either while in contact with the solid particles or after its removal from the soil by washing with distilled water, upon the physical condition of soils or complex silicates of such a character as to admit of chemical exchange of ions, is, in a measure, dependent upon the nature of the chemical

bodies formed by virtue of this interchange of ions. The soluble products, including the salts of calcium, magnesium or other bases, which are formed when salt solutions are allowed to act upon soils, must be considered, if a true conception of the physical effects of the added salts is to be secured. In addition, the individual properties of the new silicate complex, which is formed simultaneously with the exchange of ions, must be recognized as bearing significantly on the final effects of the added salt on the physical condition of the soil. Undoubtedly, these factors have a bearing on the effects of salts on the capillary movement of water in soils, and make impossible the direct application of physical laws concerning surface tension and densities to the phenomena of capillary rise, as Briggs and Lapham<sup>38</sup> have attempted. Instead, the factors discussed above may help to harmonize the observations made by these investigators with the more recent findings of Kossovich.<sup>39</sup>

The addition to soils of KCl, NH<sub>4</sub>Cl, NaNO<sub>3</sub>, Na<sub>2</sub>SO<sub>4</sub> or NaCl, and in fact most neutral salts, has been shown by Way,<sup>40</sup> Eichhorn,<sup>41</sup> Henneberg and Stohman,<sup>42</sup> Peters,<sup>43</sup> Van Bemmelen,<sup>44</sup> Bradley,<sup>45</sup> Curry and Smith,<sup>46</sup> and others, to result in increasing the quantity of calcium, magnesium and other metallic ions in the solution secured from soils so treated, as compared with the same soils receiving distilled water. Furthermore it has been shown by Hall and Morison,<sup>47</sup> Davis,<sup>48</sup> Patten and Gallagher,<sup>49</sup> Masoni,<sup>50</sup> and others that calcium and magnesium salts possess more pronounced flocculating powers than the corresponding salts of the alkali metals. This fact may account for the different rates at

<sup>38</sup> U. S. Dept. Agric., Bur. of Soils, Bull. 19, 1902.

<sup>39</sup> Cited from Exp. Sta. Rec., vol. 25, no. 9, p. 824.

<sup>40</sup> *Loc. cit.*

<sup>41</sup> Cited from Sullivan, *loc. cit.*, p. 12.

<sup>42</sup> Cited from Sullivan, *loc. cit.*, p. 13.

<sup>43</sup> Landw. Vers. Stat., vol. 2, p. 113, 1860.

<sup>44</sup> *Loc. cit.*

<sup>45</sup> Oregon Agric. Exper. Sta., Bull. 112, 1912.

<sup>46</sup> New Hampshire Agric. Exper. Sta., Bull. 170, 1914.

<sup>47</sup> *Loc. cit.*

<sup>48</sup> U. S. Dept. Agric., Bur. of Soils, Bull. 82.

<sup>49</sup> U. S. Dept. Agric., Bur. of Soils, Bull. 52, 1908.

<sup>50</sup> Abstract in Journ. Chem. Soc. London, vol. 102, no. 597, p. 677, 1912.

which the same strength of different salt solutions operates to clarify suspensions from different soils. Moreover, the addition of these salts seems to bring about a decided increase in the colloidal content of the soil, which is obviously manifested when the soluble salts are removed by distilled water. On the other hand, the introduction of calcium salts to a soil does not apparently bring about the production of a colloidal complex, although some interchange of ions occurs. In fact, the addition of  $\text{CaCl}_2$  followed with washing seems to have the opposite effect on the Davis clay-loam soil. For these reasons it seems evident that the insoluble as well as the soluble products formed by the interaction of salts on soils must be considered as important factors in such practices as prescribe the use of soluble salts on soils.

It is apparent that at least the Na, K and  $\text{NH}_4$  ions form a colloidal substance upon their introduction into the silicate complexes of the Davis soil. The Ca-ion, and probably others, form, on the contrary, non-colloidal substances when introduced into the complex silicates of the Davis soil. It is also made clear that the acid ion is of little importance in the phenomena following the washing out of salts from the soil, by the fact that  $\text{NaNO}_3$ ,  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$  all operate to produce approximately the same results.

#### SOME GENERAL OBSERVATIONS ON OTHER PECULIAR APPEARANCES ACCOMPANYING THE WASHING OUT OF CERTAIN SALTS FROM SOILS

Stewart<sup>51</sup> in a recent paper attributes the color of the nitre spots to the action of sodium or potassium nitrates on the organic matter of the soil. Bearing somewhat upon this subject, we have noted that the addition of  $\text{NaCl}$  or  $\text{Na}_2\text{SO}_4$  to a soil materially increases the organic matter in the supernatant solution if the depth of color of such a solution can be relied upon as an index of the quantity of organic matter contained therein. But the most striking results in this connection appear when these neutral salts are removed from the soil by washing. The soil itself assumes the typical gray color of alkali soils in the field, and the

<sup>51</sup> Journ. Amer. Soc. Agron., vol. 6, no. 6, p. 247, 1914.

filtrate takes on a rich, dark brown color, not unlike the usual  $\text{NH}_4\text{OH}$  humus extract. Frequently the soil when dry is covered with a thin layer of hard, black, organic matter. Indeed the resemblance between the  $\text{NaCl} + \text{H}_2\text{O}$  soil, the water in contact with it, or its percolate, and the natural alkali soils and the water bathing their particles, is so striking as to deceive even those experienced in the handling of "black alkali" soils.

The increased solubility of the organic matter in the salt-treated, water-washed soils, as indicated by the increased depth of the brown color, cannot be due to the presence of the  $\text{NaCl}$  as a chemical entity, for the color does not appear until after some of the salt has been removed from the soil by washing. There can be no doubt that the  $\text{NaNO}_3$  by means of an interchange of ions with the organic salts of calcium or magnesium does affect a greater solubility of organic matter, but this solubility may be greatly increased if the added salt is washed from the soil. This fact may be of assistance in accounting for the color of the nitre spots.

In agreement with Van Bemmelen<sup>52</sup> and Warington,<sup>53</sup> we have also noted the appearance of fluffy, flocculent colloids passing through the filter paper, when the added salt is practically washed from the soil. These colloids coagulate when in contact with the filtrate containing the soluble salts from the soil.

One other point which has not received prior attention, but which seems of sufficient importance to be mentioned at this time, is the fact that all of the salt need not be washed from the soil to produce the effects noted. In another series of experiments, which will be reported in detail later, 3200 cc. of a 1.5 per cent solution of  $\text{NaCl}$  (which was intended to represent the strength of the soil solution when 0.3 per cent of  $\text{NaCl}$  is added to the soil under optimum water conditions) was passed through 1600 grams of Davis soil. The soil was affected to a greater extent than a soil receiving a similar quantity of  $\text{NaCl}$ , namely 4.8 grams, or than a third soil receiving a similar quantity of  $\text{NaCl}$ , which in addition was leached with 3200 cc. of water. The injury to the first soil was greater than to the other soils despite the fact that

<sup>52</sup> *Loc. cit.*

<sup>53</sup> *Loc. cit.*

a small quantity of salt remained in contact with the soil. These observations lend further confirmation to the fact previously observed, that a second addition of NaCl to a  $\text{NaCl} + \text{H}_2\text{O}$  soil does not cause the reversion of the soil to its original condition.

#### GENERAL DISCUSSION OF THE EFFECTS NOTED AND THEIR APPLICATION TO SOIL STUDIES AND THE MANAGEMENT OF SOILS

In the discussion on the possible relation of the calcium and magnesium content of the percolate to the diffused condition of salt-treated, water-washed soils, attention has already been called to the fact that the phenomena, first observed in case of the cylinders containing Davis soil, also appeared in three other soils of widely different types. Evidently the effects observed when added salts are washed from soils by distilled water, are not peculiar to any particular type of soil, but are more or less general in their application. Some soils, as those which do not offer replaceable calcium or magnesium to exchange for sodium or other ions of salt solutions, would probably react to a much less extent than those soils which do react with salt solutions. The degree to which the soil is affected by the salt-and-water treatments is apparently dependent upon the amount of exchange of bases, or the direct addition of bases which occurs. This, of course, varies markedly with different soil types and is probably closely associated with complex silicates of which mention has already been made. This conception conforms, in the main, with the ideas brought out by Knop,<sup>54</sup> Van Bemmelen,<sup>55</sup> and Warington,<sup>56</sup> with respect to the absorption of salts by soils. Moreover, Kossovich<sup>57</sup> has noted that NaCl exercises a more pronounced effect on capillary rise in clay soils than in sandy soils.

There can be but little doubt that the effects of salts on the physical and chemical properties of soils has a wide range of application to alkali soils and their management. Moreover, the

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<sup>54</sup> Cited from U. S. Dept. Agric., Bur. of Soils, Bull. 52, p. 19.

<sup>55</sup> *Loc. cit.*

<sup>56</sup> *Loc. cit.*

<sup>57</sup> *Loc. cit.*

peculiar phase of these effects under consideration would seem to have particular significance in that connection. First of all, these soils contain, as a general rule, sufficiently large quantities of salts to be commensurate with those used in the experiments reported above. Secondly, the natural rainfall, irrigation practices, and drainage operations often wash the salts out of the top layers of soil, so that this soil would be exposed to the conditions likely to produce the inferior physical qualities attendant upon deflocculation.

During the late winter and early spring months the writer had occasion to observe the standing water in the depressions of the alkali lands near Fresno, California. To all appearances the underdrainage of these low areas would be practically impossible, owing to the imperviousness of the soil, which upon examination reveals all the characteristics of the diffused salt-treated, water-washed soils. Besides, the gray appearance of the soil when dry, and the striking resemblance of the supernatant liquid to that above the soils in the cylinders, would certainly lead one to believe that even under natural conditions the washing out from and the dilution of the salts in alkali soils has much in common with the artificial production of similar conditions.<sup>58</sup> Heretofore, the depressions and the remarkable imperviousness of their soils have been attributed to the presence of  $\text{Na}_2\text{CO}_3$ .

It would seem that the deflocculation effects exhibited by soils when added salts are washed from them would have particular bearing upon the reclamation of alkali soils by underdrainage. Yet the possible application of this feature to alkali reclamation seems to have been omitted from the literature dealing with this subject.<sup>58</sup> Cameron and Patten<sup>59</sup> observed somewhat similar lowering of the rate of percolation, followed, however, by an increase later as the washing out of the salts progressed. Hare,<sup>60</sup> in a recent paper describing tank experiments dealing with the effects of alkali salts on soils and crops, does not note any possible effect of the added salts on percolation through the soils receiving them, but does comment on the observation that  $\text{Na}_2\text{SO}_4$

<sup>58</sup> Hilgard, *Soils*, 1911, chap. 22, and U. S. Dept. Agric., Bur. of Soils, Bull. 35, 1906.

<sup>59</sup> *Journ. Amer. Chem. Soc.*, vol. 28, p. 1639, 1906.

<sup>60</sup> *New Mexico Agric. Exper. Sta.*, Bull. 88, 1913.

was more difficult to leach from the soil than  $\text{Na}_2\text{CO}_3$  or  $\text{NaCl}$ . Specific information on the physical condition of the drained alkali soils of the various drainage experiments is lacking. Even the recorded observations of a general nature are indefinite and discordant. Thus at Salt Lake,<sup>61</sup> Tempe,<sup>62</sup> and North Yakima,<sup>63</sup> the effect of drainage has not appeared to have benefitted the physical condition of the soil, while at Fresno,<sup>64</sup> Billings,<sup>65</sup> and elsewhere, the drained soil appeared to be superior with respect to physical condition than the undrained soil. Owing to the inadequacy of these reports in that no definite measurements of alteration in the physical condition of the soil are given, they cannot be accepted as a final expression of the effects of drainage of alkali soils.

The behavior of the Davis soil in the cylinders and the general trend of the laboratory experiments seem to the writer convincing arguments that the drainage of these cylinder soils in their present condition would be practically, perhaps absolutely, impossible. Underdrainage, supplemented with powerful flocculating agents, might serve as a feasible plan whereby the excess salt could be removed from such soils.

That natural alkali soils are not strictly comparable with the cylinder soils to which single salts have been added is patent, and this may account, in a measure, for the general discrepancy between the behavior of certain natural alkali soils and that of alkali soils made artificially by the addition of salts. The washing out of salt mixtures, as in the case of natural alkali soils, may have an entirely different effect than the washing out of a single salt. It has, however, been shown in this laboratory that the washing out of certain mixtures of salts from the Davis soil has resulted in the same way as washing out single sodium salts. On the other hand, the time for the reaction of the salt in the soil

<sup>61</sup> Dorsey, C. W., U. S. Dept. Agric., Bur. of Soils, Bull. 43, p. 16, 1907, and Bull. 35, p. 181, 1907.

<sup>62</sup> Dorsey, C. W., U. S. Dept. Agric., Bur. of Soils, Bull. 35, p. 190, 1907.

<sup>63</sup> *Loc. cit.*, p. 189.

<sup>64</sup> Mackie, W. W., U. S. Dept. Agric., Bur. of Soils, Bull. 42, p. 43, 1907.

<sup>65</sup> Dorsey, C. W., U. S. Dept. Agric., Bur. of Soils, Bull. 44, p. 18, 1902.

being comparatively unlimited in case of the natural soils, it may considerably alter the end-points and end-products and thus introduce a new factor. The constant up-and-down movement of the soluble salts in the soil, the alternate drying and wetting of the soil, and the effects of thermal changes on the soil, may partially obliterate the true effects of washing certain salts from soils. In addition, the relation of calcium, magnesium and other bases to the sodium may be modified by the conditions just mentioned, so that different equilibria are established. Furthermore, the nature of the soil, especially with respect to the original content of colloidal substances and hydrated silicate complexes, must be taken into account. Thus the effect of draining salts from sandy soils free of colloids may be so small as to pass unrecognized, while similar drainage from heavier types of soils would produce marked effects. The colloidal content, too, and the state of diffusion thereof, in our drained alkali lands, may be a direct result of the first natural introduction of the salts into and their subsequent removal from the original soil, the exact physical condition of which may be only surmised. Hence we have no means for securing evidence as to the exact salt effects under natural conditions.

As suggested by Mayer,<sup>66</sup> the physical effects resulting from washing sea salts from the soil may be as injurious to the growth of plants as the direct toxicity of the salt itself. Our experience with the cylinders serves to confirm this idea, for despite the fact that the top foot of soil is almost free from salts, it seems impossible to secure a satisfactory growth of plants. A pot experiment, to be reported upon later, also gives substantial proof that the deflocculated soil, comparatively free from salt, offers a far less congenial home for barley plants, at least during the early stages of growth, than the same soil containing as high as 0.3 per cent of NaCl.

The inability of the plants to make a satisfactory growth in the highly deflocculated soils appears, from a cursory consideration of the conditions, to be due to the lack of available moisture. Although the deflocculated soils may seem to be moist, yet the quantity of water which the plants have at their disposal is

<sup>66</sup> *Loc. cit.*



probably insufficient to promote normal growth. At least this condition is suggested by the moisture equivalents of the deflocculated  $\text{NaCl} + \text{H}_2\text{O}$  soil as determined with the centrifugal machine developed by Briggs. In addition to the low availability of the moisture in such soils as an inhibiting factor, the rate of movement of water through such soils must also be considered in its relation to the growth of plants. From all appearances, it seems justifiable to predict that the movement of water through the highly diffused soils is so slow as to fail to resupply the area occupied by the roots from the more moist soil layers lying adjacent to the root area.

The observed effects of washing salts from soils may also have some bearing upon fertilizer practices and the results on crop plants secured therefrom. The salts used as fertilizers undoubtedly effect certain modifications of the physical condition of soils. It is also true that the washing out from the soil of at least some of these salts brings a far greater change in the physical properties of the soil than that accompanying the application of the salts. Probably no instance with respect to this feature, as a phase of soil management, is so patent as that concerning the use of  $\text{NaNO}_3$ . Hall<sup>67</sup> comments upon the inadvisability of applying this material under certain conditions because of the deflocculating effect of the alkalinity due to residual sodium carbonate. Warington<sup>68</sup> also notes the deflocculating effect of  $\text{NaNO}_3$  and refers to it as being particularly noticeable after heavy rains. More recently McGeorge<sup>69</sup> has observed a marked retardation of percolation through Hawaiian soils receiving applications of  $\text{NaNO}_3$ , but attributes this effect to some reaction between the added salt and the organic matter of the soil. Undoubtedly the application of  $\text{NaNO}_3$  to soils under certain circumstances has resulted in a deterioration of the physical condition of the soil, although the  $\text{NaNO}_3$ , as a salt, is in itself a flocculating agent.

The writer has produced marked deflocculation in the Davis soil by applying  $\text{NaNO}_3$  and subsequently washing the soluble

<sup>67</sup> The Soil, p. 252, 1910.

<sup>68</sup> Loc. cit.

<sup>69</sup> Hawaiian Agric. Exper. Sta, Bull. 35, 1914.

salts from the soil with water. The use of the  $\text{NO}_3$  radical by plants or bacteria, thereby leaving the sodium, could not have occurred to any extent in this case, for the whole process required no longer than two hours, and was carried out in a filter. The filtrate at no period in the washing process showed sufficient alkalinity to account for the deflocculation. Hence the former conception that residual  $\text{Na}_2\text{CO}_3$  causes the deflocculation seems untenable, and some such hypothesis as has been advanced in this paper must be adopted to explain the unfavorable physical condition frequently existing in soils receiving applications of  $\text{NaNO}_3$ .

With respect to fertilizer salts in general, Hall<sup>70</sup> calls attention to the deflocculation following the use of neutral salts on soils, but believes the effect is due to alkalinity arising from the absorbed base. Hessler<sup>71</sup> has noted the increase in coherence of the soil particles when  $\text{NaCl}$ ,  $\text{NaNO}_3$  and kainite have been applied to soils. Hoffman,<sup>72</sup> however, could not detect any difference in the interior surface of soils due to fertilizer applications of the usual magnitude. It is of interest to note that no attempt is made to correlate the inferior physical condition of salt-treated soils with the process of leaching the salts from them.

However, potassium and ammonium salts are effective in the same direction as sodium salts when washed from the soil, although they are generally considered flocculating agents when in contact with certain colloidal particles. Hence, to be fully comprehensive and expressive of the whole truth, studies on fertilizer effects should involve not only the conditions under which the salt or salts are present, but also those conditions which not infrequently arise in nature, whereby the soluble salts are removed from the soil by processes analogous to washing the soil with water.

The nature of the clay colloids and the cause of the relatively high degree of deflocculation which they assume when suspended in water have long been subjects of much conjecture and much

<sup>70</sup> *Loc. cit.*

<sup>71</sup> Cited from *Exper. Sta. Rec.*, vol. 31, no. 2, p. 123, 1914.

<sup>72</sup> *Landw. Vers. Stat.*, vol. 85, nos. 1-2, p. 123, 1914.

debate. Hilgard<sup>73</sup> has consistently maintained that the power of such colloids to remain in a stabilized condition is not to be attributed solely to the fineness of division of such particles, but that other factors may be responsible for that condition. In agreement therewith, our experience, which has been presented in this communication, has led us to believe that the chemical nature of the body itself and that of the medium, determine in a large measure the condition in which the colloid may exist.

The extent to which the factor considered in this paper may be applicable to agricultural practice can be better surmised than asserted at the present writing, but it seems highly possible that the modifications of the physical condition of the soil, due to washing out the soluble salts, under circumstances involving fertilizer applications or natural alkali soils, will be reflected in the inferior tilling qualities of the soil, in the increased resistance offered by such soils to root penetration, in the lack of air space and air movements, in the deflocculated soil, and in the moisture and temperature relations of such diffused soils. The movement of moisture in soils by surface tension and osmotic pressure or under gravitational attraction, appears to be particularly dependent upon the degree of deflocculation of the soil colloids.

Besides affecting the physical condition of the soil, the leaching out of soluble neutral salts from soils is, as has been previously shown, frequently accompanied by notable quantities of calcium and magnesium in the percolate. Thereby the soil sustains a considerable loss of calcium and magnesium, which may in the course of time be of sufficient magnitude actually to deplete the available supply of these plant-food elements in the soil. The bacterial flora and bacterial activity of soils subjected to the treatments outlined above are liable to be considerably modified and to all appearances in a harmful direction. Thus all the factors of soil fertility are likely to be measureably affected through the process of washing salts from soils; consequently the crop-producing power of the salt-treated, water-washed soil is apt to be considerably modified.

<sup>73</sup> Soils, chap. VI.

## SUMMARY

1. The Davis clay-loam soil to which surface applications of solutions of  $\text{NaCl}$ ,  $\text{Na}_2\text{SO}_4$ , and  $\text{Na}_2\text{CO}_3$  had been made became very impervious to water, difficult to cultivate, and manifested the characteristics of a high degree of diffusion, although these salts have been shown to exercise flocculating powers on suspensions of this soil.

2. The salt-treated soils referred to were in cylinders exposed to natural conditions.

3. Examination of the soils showed that the salts had moved downward into the lower layers of soil and that only the surface soil had been affected in the direction described.

4. The deflocculated condition resulting from adding certain salts to and subsequently washing them from soils can be reproduced in the laboratory.

5. The deflocculation of soils treated in the manner described above is intimately associated with the leaching of the  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$  down into the lower layers of soil by water. In the case of  $\text{Na}_2\text{CO}_3$  the leaching process is not so essential for the diffusion of the soil colloids.

6. The addition of  $\text{NaCl}$ ,  $\text{Na}_2\text{SO}_4$ , and  $\text{Na}_2\text{CO}_3$  to the Davis soil when followed with applications of water was particularly effective in diminishing the rate of percolation through the soil so treated.

7.  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$  in constant contact with the Davis soil increased the rate of percolation, except when a comparatively dilute solution of  $\text{NaCl}$  was slowly passed through the soil for a considerable period of time.

8. The Davis soil treated with  $\text{NaCl}$ ,  $\text{NaOH}$ ,  $\text{Na}_2\text{CO}_3$  and other salts, followed by leaching with water, yields a suspension in water containing approximately ten times as much solid matter as the same soil washed with water only. A real diffusion in such salt-treated soils seems evident.

9. The soil once diffused by washing out added  $\text{NaCl}$  requires considerably more salt to completely flocculate it, than does the water-washed soil. Likewise the injured physical condition of such soils is not readily repaired by a second addition of  $\text{NaCl}$ .

10. The portion of the organic matter of the soil known as humus has little or no connection with the appearance of diffusion in salt-treated, water-washed soils.

11. The diffusion in soils treated as described above seems to be closely associated with the direct addition of sodium to, or with the absorption of sodium by the soil, thereby producing a new silicate complex of a colloidal character in the soil.

12. This silicate complex is formed simultaneously with the interchange of ions occurring between the salt and the soil.

13. The washing process serves, in the case of neutral salts, to remove flocculating agents.

14. The loss of calcium and magnesium from the soil bears little or no relation to the flocculation appearing in salt-treated, water-washed soils except in so far as it may be a measure of the absorbed sodium.

15. The presence of the OH-ion does not seem to be an essential factor in the diffusion of salt-treated, water-washed soils.

16.  $\text{Na}_2\text{CO}_3$  and NaOH produce markedly different effects on suspensions of the Davis soil.

17. The acid ion of the salt is not an important factor in the deflocculation phenomena following the washing out of salts from soils.

18. Sodium, potassium, and ammonium seem to produce the colloidal silicate complex when salts of these metals are applied to soils, while calcium does not.

19. Dilute solutions of acids and salts possess flocculating powers on suspensions of the Davis soil.

20. It is not essential in every case to wash all of the salt out in order to bring about diffusion.

21. The facts presented are discussed in their relation to the reclamation and management of alkali lands, and in their application to the use of soluble salts as fertilizers.

The writer desires to express his thanks to Professor Charles B. Lipman for many valuable suggestions and for his critical reading of the manuscript, and to L. E. Bailey for his timely assistance in the analytical work.

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INFLUENCE OF THE COMPOSITION AND  
CONCENTRATION OF THE NUTRIENT  
SOLUTION ON PLANTS GROWN  
IN SAND CULTURES

BY  
ARTHUR HUGO AYRES

UNIVERSITY OF CALIFORNIA PRESS  
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INTRODUCTION

Since the recognition of the fact that the mineral content of the plant body is derived from the mineral constituents of the soil, the part which the soil solution takes in the nutrition of the plant has been the subject of numerous investigations by chemists, plant physiologists, and soil scientists, who have made large contributions to our knowledge in this important field. The early investigations of Knop and other plant physiologists showed conclusively that the elements which are essential to plant growth are carbon, hydrogen, oxygen, nitrogen, sulphur, phosphorus, potassium, magnesium, calcium, and iron. As most of this work was done before the development of the new chemical and physical theories, in regard to solutions in particular, the problems dealing with the absorption of these elements remain for explanation in the light of this new knowledge. The modern period of research in this field has thus been characterized by an intensive study of the absorption of nutrient elements by the plant. The earlier conceptions, which had a marked tendency to link each element with some specific physiological process or with the development of some morphological part of the plant, have been



largely discarded in view of the recent investigations, which have shown that plant growth is not a simple function of any any particular element, but is to a very large extent influenced by the combinations of elements in the solution from which the plant derives its nourishment. Thus while calcium may in certain cases act as a neutralizer of oxalic acid,<sup>1</sup> it exhibits a more general function of antagonism for salts of potassium, sodium, magnesium, and other salts which would be toxic if calcium were not present.<sup>2</sup> While this antagonistic function may be characteristic, it would seem from the experiments of Tottingham<sup>3</sup> that either this antagonistic action of calcium or the toxic effects of magnesium are influenced by the total concentration of the solution. Tottingham, therefore, concludes that the injurious effects of magnesium depend not only on the amount of calcium present but also upon the complex balance between all the salts in the solution. It follows that while the later investigations have undoubtedly given a wider conception of the rôle of the various nutrient elements, the exact relation which exists between the recognized nutrient function of these elements and the balancing function in the solution is not definitely known.

The part which the total concentration of the solution takes in the complicated problem of plant nutrition is by no means clear. On the one hand, the experimental evidence of Cameron<sup>4</sup> and his co-workers shows that the plant growing in water culture is not influenced to any extent by wide variations in the total concentration of the solution, a view which is further supported by the researches of Tottingham,<sup>5</sup> who concludes that nutrient solutions ranging from 0.01 per cent to 0.14 per cent do not affect the dry weight in the case of wheat grown in these solutions. A similar view is taken by Stiles,<sup>6</sup> who thinks that the individual variation of plants grown in water cultures is as large or larger than that which is often accredited to a variation in the compo-

<sup>1</sup> Schimper, *Flora*, vol. 73, pp. 207-261, 1890.

<sup>2</sup> Loew, *Flora*, vol. 75, pp. 368-394, 1892; and U. S. Dept. Agric. Bur. Plant Ind. Bull. 45, 1903; Osterhout, *Bot. Gaz.*, vol. 42, pp. 127-134, 1906, and vol. 44, pp. 259-272, 1907.

<sup>3</sup> Tottingham, *Physiol. Res.*, vol. 1, pp. 133-245, 1914.

<sup>4</sup> Cameron, *Jour. Phys. Chem.*, vol. 14, p. 320, 1910.

<sup>5</sup> *Loc. cit.*

<sup>6</sup> Stiles, *Ann. Bot.*, vol. 29, pp. 89-96, 1915.

sition of the solution. In decided contrast to the evidence cited above, we have the investigations of Hall, Brenchley, and Underwood,<sup>7</sup> who conclude, on the basis of their results with barley plants grown in a standard solution of four different concentrations, that "the growth made by plants in the soil solution is in the main determined by the amount of plant food they contain," and that "the concentration of the nutrient solution within certain wide limits, irrespective of the total amount of the plant food available, is a factor in the rate of plant growth which varies directly though not proportionally with the strength of the solution in the particular nutrient or nutrients limiting the growth." The experiments of Shive<sup>8</sup> are of considerable interest in this connection. Wheat was grown in solutions of three different total concentrations, 0.1, 1.75, and 4.00 atmospheres, in terms of possible osmotic pressure. The solution used was the three-salt solution first used by Birner and Lucanus<sup>9</sup> containing  $\text{Ca}(\text{NO}_3)_2$ ,  $\text{MgSO}_4$ , and  $\text{KH}_2\text{PO}_4$ . All possible sets of proportion of these salts were included for increments of change equal to one-tenth of the total possible osmotic pressure. As judged from the extensive quantitative data collected, Shive concludes that the growth of wheat plants in solutions of any given salt proportion is determined by the concentration of the medium.

In the course of some experimental work concerning the dropping of flowers by  $F_1$  species-hybrids of *Nicotiana*<sup>10</sup> it seemed desirable to grow a considerable number of plants in sand cultures which would vary widely both as to the principal nutrient elements, nitrogen, phosphorus, and potassium, and as to the total concentration of all of the nutrient salts. The marked influence of the nutrient factors upon the growth of the plant has afforded an excellent opportunity for a somewhat detailed study of the influence of the composition and concentration of the nutrient solution upon the growth of one of the higher seed-plants of herbaceous character.

<sup>7</sup> Hall, Brenchley, Underwood, Jour. Agric. Sci., vol. 6, pp. 278-301, 1914.

<sup>8</sup> Shive, Physiol. Res., vol. 1, pp. 327-396, 1915.

<sup>9</sup> Birner and Lucanus, Landw. Versuchsstat., vol. 8, pp. 128-177, 1886.

<sup>10</sup> Goodspeed and Ayres, Univ. Calif. Publ. Bot., vol. 5, no. 9, 1916.

## THE USE OF SAND AS A CULTURE MEDIUM

The ease with which solutions can be prepared and subsequently analyzed has made the water-culture method especially desirable in investigations of the rôle of nutrient substances in plant growth. A solution has generally been recognized, however, to be otherwise undesirable as a medium for the growth of the higher plants, since the root system is kept, during the course of the experiment, in an unnatural environment. Thus, while this method serves admirably for analytical purposes, it seems probable that the plant thus subjected to an unnatural environment will suffer certain more or less serious physiological disturbances. In this connection, it is a well-established fact that the development of root hairs is much greater in sand than in water,<sup>11</sup> the resistance of the substratum favoring root-hair production.<sup>12</sup> Roots in general grow longer and thinner in water than in sand or moist soil. Hall, Brenchley, and Underwood<sup>13</sup> think that the more vigorous growth of barley in sand, as compared with water cultures, is due to more efficient aeration of the former. It would seem, therefore, that sand is preferable to water as a culture medium, since in sand cultures the physical conditions present about the root system more nearly simulate those found in the soil. It is still a question just what part these physical conditions may have in plant nutrition. Undoubtedly such physical reactions as capillarity<sup>14</sup> and adsorption<sup>15</sup> must be important factors, since both the absorption and availability of nutrient salts would be affected by these physical phenomena. Breazeale<sup>16</sup> has shown that the effect of concentration in sand cultures is very different from that in water cultures, the best concentration for wheat in water being three hundred parts per million, while in sand it is in the vicinity of two thousand five hundred parts per million, an effect which is no doubt largely due to the adsorption of certain salts or ions by the sand particles.

<sup>11</sup> Schwarz, *Bot. Inst. Tübingen*, vol. 1, pp. 125-188, 1883.

<sup>12</sup> Snow, *Bot. Gaz.*, vol. 40, pp. 12-43, 1905.

<sup>13</sup> *Loc. cit.*

<sup>14</sup> Bell and Cameron, *Jour. Phys. Chem.*, vol. 10, p. 659, 1906.

<sup>15</sup> Schreiner and Failyer, *U. S. Dept. Agric. Bull.* 32, 1906.

<sup>16</sup> Breazeale, *Science*, n. s., vol. 22, pp. 146-149, 1905.

The physical effects of sand as a medium for plant growth have been shown recently in a striking manner by McCall,<sup>17</sup> who added to sand the solutions which Shive<sup>18</sup> used as water cultures, with the result that much larger quantities of the nutrient salts were required than when the same species of plant was grown in water culture. The sand-culture method was selected for the present work with the above evidence in mind, and also because the growing period of the tobacco plant is long and this method precluded the tedious changing of solutions necessary when the water-culture method was used.

### EXPERIMENTAL MATERIAL

The plant which was used in the present series of investigations was a tobacco of hybrid origin derived from a cross between *Nicotiana sylvestris* (U. C. B. G. 69/09) and *Nicotiana tabacum* var. *macrophylla* (U. C. B. G. 22/07) and known in the University of California Botanical Garden as U. C. B. G. H38.<sup>19</sup> The plants to be used were raised from seed and were transferred as seedlings to the sand from the flats in which they were grown after carefully washing the roots free from adhering soil particles. In picking out seedlings from the flats care was taken to choose from the large number of plants available only those which were most nearly uniform with reference to size and general appearance. The plants were kept during the experiment in a well-ventilated greenhouse.

The sand used was a light-colored beach sand which on analysis by means of the acid-digestion method of Hilgard showed the following composition:

Fe <sub>2</sub> O <sub>3</sub>	0.592 per cent	P <sub>2</sub> O <sub>5</sub>	0.004 per cent
Al <sub>2</sub> O <sub>3</sub>	0.46 per cent	MgO	0.28 per cent
K <sub>2</sub> O	Trace	CaO	0.06 per cent

The water-holding capacity was 22 per cent when saturated. The sand was prepared for the experiment by washing in a heavy stream of tap water which was allowed to percolate through

<sup>17</sup> Unpublished work.

<sup>18</sup> *Loc. cit.*

<sup>19</sup> Setchell, Univ. Calif. Publ. Bot., vol. 5, pp. 1-86, 1912.

a column of the sand for a period of twenty-four hours. The excess of tap water retained by the sand was in turn washed out with distilled water. While this sand is inferior to the best grade of pure quartz sand in freedom from inorganic material, it is unable to supply available nutrient elements in sufficient quantity to cause any perceptible increase in growth (pl. 14). Two thousand gram portions of the sand treated as above were weighed into six-inch flower-pots which had been previously prepared by dipping them into melted paraffin. The paraffining effectually closed the pores and prevented the absorption of the culture solution by the pot.

#### DISTRIBUTION OF NUTRIENT SALTS

The pots were divided into three different groups designated as series I, II, and III, each series consisting of twenty pots divided into groups of five pots each, and a duplicate in each case, making forty pots in all in each series. In series I, nitrogen, as  $\text{NaNO}_3$ , was the varying factor within each group, the weight of each of the other salts being held constant. Thus, in series I, the pots 1 to 5 in each of the four groups contained  $\text{NaNO}_3$  as follows: 0.02, 0.2, 1.0, 2.0, 3.0 grams. In a similar manner phosphorus and calcium, as  $\text{Ca}(\text{H}_2\text{PO}_4)_2$ , were varying factors in series II, while in series III potassium, as  $\text{K}_2\text{SO}_4$ , was varied. As noted above, the twenty plants of each series were divided into four groups designated respectively as A, B, C, and D, each group consisting of five plants and a control for each. The weight of one varying factor remained the same in pots of like number through all four groups. Thus plants I A 1, I B 1, I C 1, and I D 1 each contained 0.02 grams of  $\text{NaNO}_3$ , and I A 2, I B 2, I C 2, and I D 2 contained 0.2 grams of  $\text{NaNO}_3$ . But from group A to D the weight of the other nutrient factors decreases, so that group D contains two-thirds, C one-half, and B one-fourth the weight of each of these nutrient factors as present in the A group. The effect of this distribution of salts is to give at least three important variables. First, the single nutrient salt in increasing proportions from plant 1 to plant 5 in each group, and second, the factor of total concentration which decreases from group A

to group D in each series. The third important variant, the balance of salts in the solution, follows as a matter of necessity, since the single nutrient salt added in increasing quantities from plant 1 to plant 5 is added with uniform variation and in the same quantity in each group, while the quantities of the other salts, although constant within a group, are not constant within all

TABLE 1

WEIGHT IN GRAMS OF SALTS ADDED TO EACH POT CONTAINING 2000 GRAMS OF SAND

(In the following table the roman numerals indicate the series numbers, and the letters are those of the corresponding groups.)

Pot No.	NaNO <sub>3</sub> , grams	Ca (H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> ·H <sub>2</sub> O, grams	K <sub>2</sub> SO <sub>4</sub> , grams	MgSO <sub>4</sub> ·7 H <sub>2</sub> O, grams	Total concen- tration, grams	Pot No	NaNO <sub>3</sub> , grams	Ca (H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> ·H <sub>2</sub> O, grams	K <sub>2</sub> SO <sub>4</sub> , grams	MgSO <sub>4</sub> ·7 H <sub>2</sub> O, grams	Total concen- tration, grams
IA1	.02	1.2	2.4	.96	4.58	IIC1	1.5	.02	1.2	.48	3.20
IA2	.2	1.2	2.4	.96	4.76	IIC2	1.5	.10	1.2	.48	3.28
IA3	1.0	1.2	2.4	.96	5.56	IIC3	1.5	.20	1.2	.48	3.38
IA4	2.0	1.2	2.4	.96	6.56	IIC4	1.5	1.0	1.2	.48	4.18
IA5	3.0	1.2	2.4	.96	7.56	IIC5	1.5	2.0	1.2	.48	5.18
IB1	.02	.8	1.6	.64	3.06	IID1	.75	.02	.6	.24	1.61
IB2	.2	.8	1.6	.64	3.24	IID2	.75	.1	.6	.24	1.69
IB3	1.0	.8	1.6	.64	4.04	IID3	.75	.2	.6	.24	1.79
IB4	2.0	.8	1.6	.64	5.04	IID4	.75	1.0	.6	.24	2.59
IB5	3.0	.8	1.6	.64	6.04	IID5	.75	2.0	.6	.24	3.59
IC1	.02	.6	1.2	.48	2.30	IIIA1	3.0	1.2	.02	.96	5.18
IC2	.2	.6	1.2	.48	2.48	IIIA2	3.0	1.2	.10	.96	5.26
IC3	1.0	.6	1.2	.48	3.28	IIIA3	3.0	1.2	.20	.96	5.36
IC4	2.0	.6	1.2	.48	4.28	IIIA4	3.0	1.2	1.0	.96	6.16
IC5	3.0	.6	1.2	.48	5.28	IIIA5	3.0	1.2	2.0	.96	7.16
ID1	.02	.3	.6	.24	1.16	IIIB1	2.25	.8	.02	.64	3.71
ID2	.2	.3	.6	.24	1.34	IIIB2	2.25	.8	.10	.64	3.79
ID3	1.0	.3	.6	.24	2.14	IIIB3	2.25	.8	.20	.64	3.89
ID4	2.0	.3	.6	.24	3.14	IIIB4	2.25	.8	1.0	.64	4.69
ID5	3.0	.3	.6	.24	4.14	IIIB5	2.25	.8	2.0	.64	5.69
IIA1	3.0	.02	2.4	.96	6.38	IIIC1	1.5	.6	.02	.48	2.60
IIA2	3.0	.10	2.4	.96	6.46	IIIC2	1.5	.6	.10	.48	2.68
IIA3	3.0	.20	2.4	.96	6.56	IIIC3	1.5	.6	.20	.48	2.78
IIA4	3.0	1.0	2.4	.96	7.36	IIIC4	1.5	.6	1.0	.48	3.58
IIA5	3.0	2.0	2.4	.96	8.36	IIIC5	1.5	.6	2.0	.48	4.58
IIIB1	2.25	.02	1.6	.64	4.51	IIID1	.75	.3	.02	.24	1.31
IIIB2	2.25	.10	1.6	.64	4.59	IIID2	.75	.3	.10	.24	1.39
IIIB3	2.25	.20	1.6	.64	4.69	IIID3	.75	.3	.20	.24	1.49
IIIB4	2.25	1.0	1.6	.64	5.49	IIID4	.75	.3	1.0	.24	2.29
IIIB5	2.25	2.0	1.6	.64	6.49	IIID5	.75	.3	2.0	.24	3.29

the groups of a series. Thus I A 5, I B 5, I C 5, and I D 5 each contained 3 grams  $\text{NaNO}_3$ , but the content of each pot in  $\text{K}_2\text{SO}_4$  was 2.4, 1.6, 1.2, and 0.6 grams, respectively; hence the balance between  $\text{NaNO}_3$  and  $\text{K}_2\text{SO}_4$  is very different in each of the four pots. Table 1 shows the distribution of the salts in the three series and the total concentration in grams of the salts in each pot. The total quantity of nutrient salts was added to the sand at the beginning of the experiment.

The plants were placed in the sand June 15 and were harvested about November 1, the tops and roots being kept for the determination of dry weight.<sup>20</sup> It will be seen that the plant was allowed sufficient time to complete its natural period of growth, thus permitting certain observations of a quantitative nature as recorded in table 2.

## PHYSIOLOGICAL EFFECT OF NITROGEN

### DISTRIBUTION OF SALTS IN SERIES I

In series I nitrogen as  $\text{NaNO}_3$  was the salt which was used in the same weights in all of the four groups A, B, C, and D, being present in pots 1 to 5 as follows: 0.01 g. (.001 per cent), 0.2 g. (.01 per cent), 1.0 g. (.05 per cent), 2.0 g. (.1 per cent), 3.9 g. (.15 per cent). The maximum weights of other salts were  $\text{Ca}(\text{H}_2\text{PO}_4)_2$ , 1.2 g.;  $\text{K}_2\text{SO}_4$ , 2.4 g.;  $\text{MgSO}_4$ , 0.96 g., in group A, while the minimum quantity of these salts as added in group D was 0.3 g., 0.6 g., and 0.24 g., respectively. The total concentration in this series ranged from 1.16 g. in I D 1 to 7.56 g. in I A 5.

### HEIGHT IN SERIES I

The height of each plant was measured at two different periods. The first measurement was made when the majority of the plants were just beginning to show the first signs of flower-bud formation. The second measurement was taken five or six weeks later, when most of the plants were in full bloom. The final height of the five plants in each group is shown graphically

<sup>20</sup> An accident to the roots while drying prevented the collection of further data on their dry weight.

TABLE 2

## SUMMARY OF QUALITATIVE AND QUANTITATIVE DATA FOR EACH PLANT

(In the following table the roman numerals indicate the series numbers, and the letters are those of the corresponding groups.)

Pot No.	Height, cm.	Leaf length, cm.	Leaf width, cm.	Number leaves	Number flowers	Dry weight tops, gms.
IA1	2.5	7.6	3.7	6.	0.	1.12
IA2	4.5	9.6	4.7	7.	0.	1.60
IA3	8.	12.1	6.2	10.5	0.	1.56
IA4	57.5	13.8	9.5	17.	25.5	.....
IA5	45.5	20.8	10.2	17.5	24.	7.83
IB1	5.5	9.5	4.9	7.5	0.	1.29
IB2	3.5	8.6	4.1	7.	0.	1.40
IB3	8.	12.1	5.5	9.	1.	2.17
IB4	78.	18.8	9.5	17.5	29.	8.57
IB5	49.	19.9	10.1	17.5	21.	7.96
IC1	3.5	8.8	4.3	6.5	0.	.74
IC2	2.5	8.3	4.3	6.	0.	.79
IC3	4.5	11.2	5.6	7.	0.	1.65
IC4	94.	19.5	9.7	17.15	31.5	.....
IC5	74.	18.7	9.1	18.	29.	9.24
ID1	6.5	10.5	5.4	7.	0.	1.65
ID2	17.5	12.7	6.3	9.5	0.	1.53
ID3	59.5	15.8	7.8	15.	8.5	5.86
ID4	91.5	14.6	7.7	19.	34.5	11.12
ID5	66.5	17.3	8.6	18.5	21.5	7.60
IIA1	15.	12.9	6.4	12.	0.	2.09
IIA2	55.	18.5	8.4	15.5	14.5	6.21
IIA3	69.5	18.9	9.4	16.5	22.5	8.75
IIA4	87.	19.2	9.7	16.5	25.5	8.58
IIA5	73.	18.9	9.3	16.5	29.	8.60
IIB1	21.	13.1	6.1	12.	0.	2.71
IIB2	35.5	15.3	7.3	14.5	4.5	3.82
IIB3	43.	15.	7.4	14.	9.5	4.1
IIB4	97.	18.3	9.3	18.5	37.	9.3
IIB5	74.5	18.6	9.6	17.	23.5	7.76
IIC1	13.5	12.2	5.9	11.	0.	2.73
IIC2	27.5	13.9	6.6	12.	3.	3.22
IIC3	101.	16.5	8.2	17.5	34.5	8.20
IIC4	90.	17.3	8.9	17.	32.	8.66
IIC5	78.	17.1	8.3	18.	26.5	7.88
IID1	18.	12.8	6.	11.5	2.5	2.47
IID2	64.5	14.8	7.4	15.5	15.	5.83
IID3	74.	15.5	7.6	16.	19.5	6.27
IID4	83.5	14.2	7.5	17.5	28.	7.60
IID5	86.	13.9	7.2	17.	19.	6.75
IIIA1	56.5	16.7	8.0	14.5	6.5	4.65
IIIA2	82.5	20.	10.1	16.5	27.	7.88
IIIA3	62.	20.7	9.6	17.5	14.	6.97
IIIA4	74.	20.2	10.	17.	20.5	10.53
IIIA5	63.	22.1	10.5	17.	24.	8.11



TABLE 2—(Continued)

Pot No.	Height, cm.	Leaf length, cm.	Leaf width, cm.	Number leaves	Number flowers	Dry weight tops, gms.
IIIB1	81.5	19.9	10.1	17.	22.	8.18
IIIB2	87.	20.3	10.	17.5	24.	5.87
IIIB3	94.5	18.3	9.4	17.5	31.	8.4
IIIB4	83.	19.1	9.7	18.	26.5	4.86
IIIB5	77.5	18.8	9.7	17.	39.5	8.42
IIIC1	89.	18.	9.	17.	18.	6.58
IIIC2	102.	17.7	8.8	17.5	22.5	8.57
IIIC3	109.	17.4	8.9	18.	23.	9.4
IIIC4	88.5	18.5	9.2	17.5	25.	7.66
IIIC5	79.	17.2	8.8	18.	26.5	8.68
IIID1	89.	16.4	8.2	17.	14.	5.6
IIID2	103.5	15.1	7.8	18.	17.5	7.27
IIID3	100.5	15.9	7.8	17.5	16.5	6.96
IIID4	90.	16.5	8.2	17.5	23.	6.75
IIID5	66.5	17.5	8.7	17.5	19.	6.36

in figure 1. As two plants were given similar treatment in each case, the height as noted in table 2 is the mean of the height measurements of these two plants.

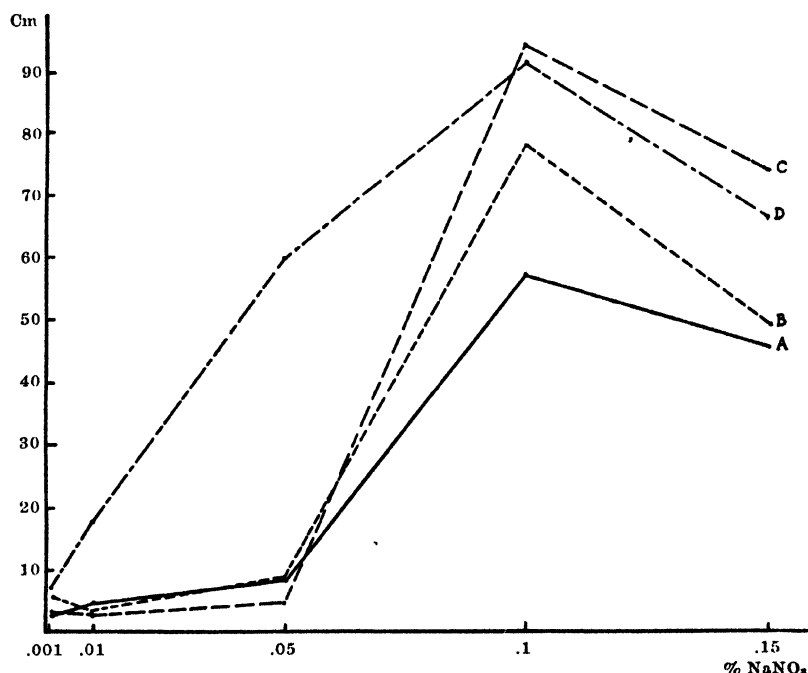


Fig. 1.—Graph showing the influence upon height of equal quantities of  $\text{NaNO}_3$  in the different groups of series I.

The effect of increasing the quantity of nitrogen from plant 1 to plant 4 is marked in all groups. In every group plant 5 is not so tall as plant 4 (pl. 5), hence it would seem that the optimum nitrogen supply for tobacco growing in sand cultures, with height as an index, is somewhere near 0.1 per cent of  $\text{NaNO}_3$ , calculated on the basis of the dry weight of the sand.

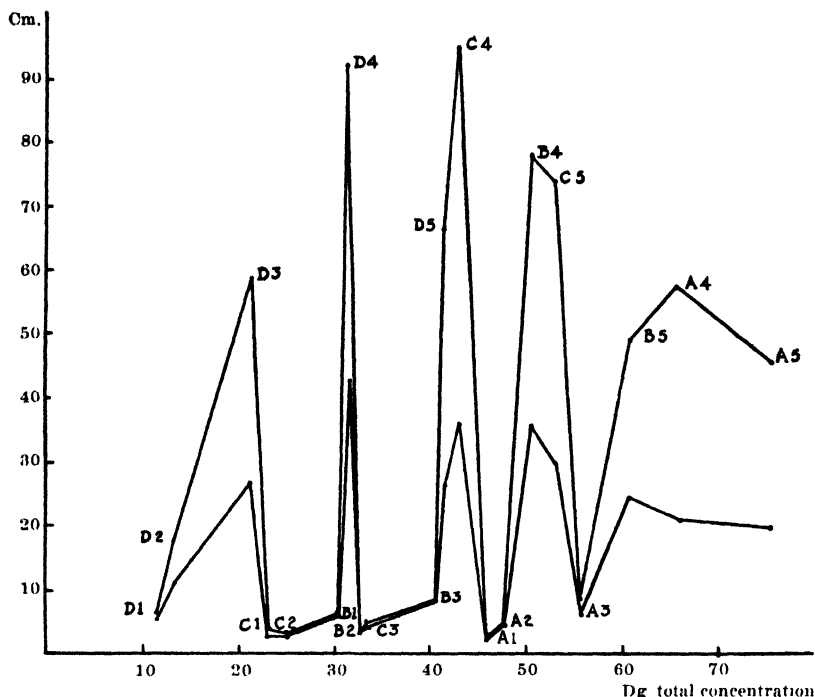


Fig. 2.—Graph showing the influence of total concentration upon height in series I.

There is slight variation in the first three plants of groups A, B, and C, showing that change in the total concentration of the solution within these limits does not affect the characters of the plants to any great extent. This, however, does not apply to group D, in which the total concentration is low (1.16 to 4.14 g.), for plants 2 and 3 are far superior to the corresponding plants of the other groups, although the nitrogen content is exactly the same. When the nitrogen content is at or near the

optimum the influence of concentration on height is exhibited in a more uniform manner, as shown in plants 4 and 5 of all groups (pl. 6). Plants growing in the solution of lower concentration, in general, are taller than those growing in the solutions of higher concentration.

The influence of total concentration upon height of plant when taken in connection with optimum and deficient nitrogen supply is indicated in figure 2, which shows a curve for each of the two measurements made on each plant, the upper curve being plotted from the last measurements taken. This curve is especially interesting since it shows the influence of nitrogen as a prime factor in growth in solutions which are of approximately the same concentration. Thus in I D 4 and I B 1, where the total concentrations are 3.14 g. and 3.06 g., the heights of the two plants are 91.5 cm. and 5.5 cm. Such a large difference can be due in this case only to the lack of nitrogen in I B 1, since there is present a considerable excess of the other nutrients required (pl. 7).

A study of the early and late height curves shows that the plant responded very early to the amount of salts available, as is indicated by the similarity of the early curve to that based upon the later measurements. The greatest final height is attained, as was to be expected, through the continuous growth of those plants which had sufficient nitrogen, while the growth of the under-nourished plant was decidedly retarded. Thus at the end of the growing season the differences in height between the two groups became more marked as the time of complete maturity drew near. Attention is called to this fact since it is not an uncommon practice in growing plants in water cultures to harvest them before maturity. While these plants might give an index of the influence of the culture solution on growth, they would not give a true value for the nutritive function of the solution, since increase might persist for a considerable time in solutions of optimum nutritive value while plants growing in an unfavorable medium would be practically at a standstill.

### DRY WEIGHT IN SERIES I

A comparison of the curves for height (fig. 1) and for dry weight (fig. 3) shows that in a general way height is an index to the dry weight. This fact is more marked in this series than in either series II or III since the plants were more uniform throughout the series, being uniformly stocky where the height was above 40 cm. In the other groups some of the plants were

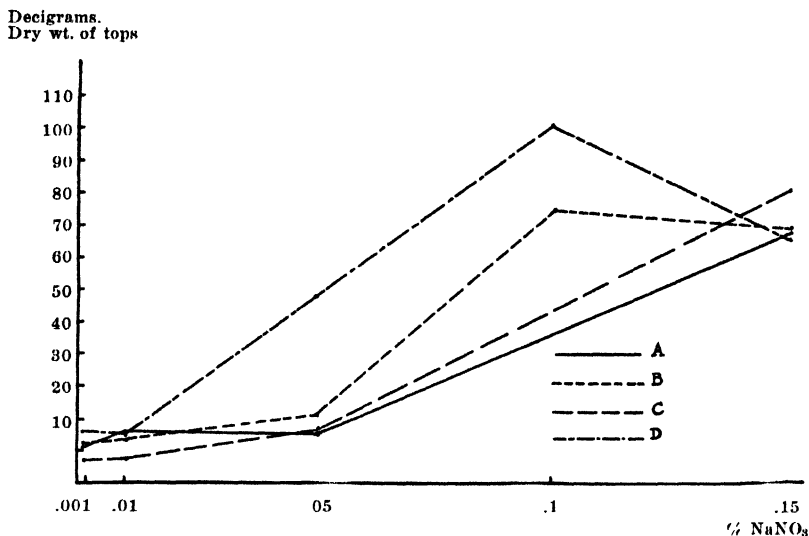


Fig. 3.—Graph showing influence of equal quantities of NaNO<sub>3</sub> in the different groups of series I upon dry weight of tops.

tall and very spindly (pl. 15), and there was a marked decrease in the length and width of leaf, which would, of course, lower the dry weight. The height measurements even on a plant of the habit of the tobacco cannot be taken alone as an indication of the nutritive value of a solution, a fact which has been shown to hold true to a much more noticeable extent in such plants as wheat, which will stool more in some cultures than in others without perceptible differences in height.

The effect of the concentration is evidenced in the curve for group D, which is much higher for plants 3 and 4 than for plants of like number in any other groups. Had it been possible to take

the dry weight for plant I C 4 this weight would probably have been slightly greater than that of I D 4 (see table 2), making the curves correspond to those for height.

#### THE INFLUENCE OF THE CULTURE SOLUTION ON FLOWER PRODUCTION

Our information concerning factors which may influence flower formation is very incomplete. Vochting<sup>21</sup> has demonstrated that light intensity is a factor in flower development in *Mimulus*, and numerous other plant physiologists have found that a more or less marked influence on flower formation could be correlated with some external factor. The relation to the formation of flowers of nutrient salts in the medium in which growth takes place seems never to have been studied in any thoroughgoing manner. Mobius<sup>22</sup> found that certain *Gramineae* flowered better on dry soil and on soil low in nutrient elements than on soil rich in nutrients and where water supply was abundant. Jost<sup>23</sup> thinks that the fact that root pruning increases flower production may be thus explained, since there is a lowering of the absorptive capacity of the tree for inorganic salts. There has been also a general opinion, which is not fully justified, that any condition which will cause marked vegetative development will retard flower production. The experiment herein reported has presented an opportunity for a study of the influence of the composition and concentration of the solution upon flower production.

In the plant used in this experiment the flowers fall soon after opening, leaving a scar upon the inflorescence stock.<sup>24</sup> At the end of the growing season the number of flowers produced by each plant was determined by counting these scars (see table 2). The total number of flowers produced by each plant is shown diagrammatically in figure 4.

<sup>21</sup> Vochting, Jahrb. f. wiss. Bot., vol. 25, p. 149, 1893.

<sup>22</sup> Mobius, Beitr. z. Lehre v. d. Fortpflanzung d. Gewachse, Jena, 1897.

<sup>23</sup> Jost, Lectures on plant physiology, p. 364, 1907.

<sup>24</sup> Goodspeed and Ayres, Univ. Calif. Publ. Bot., vol. 5, no. 9, 1916.

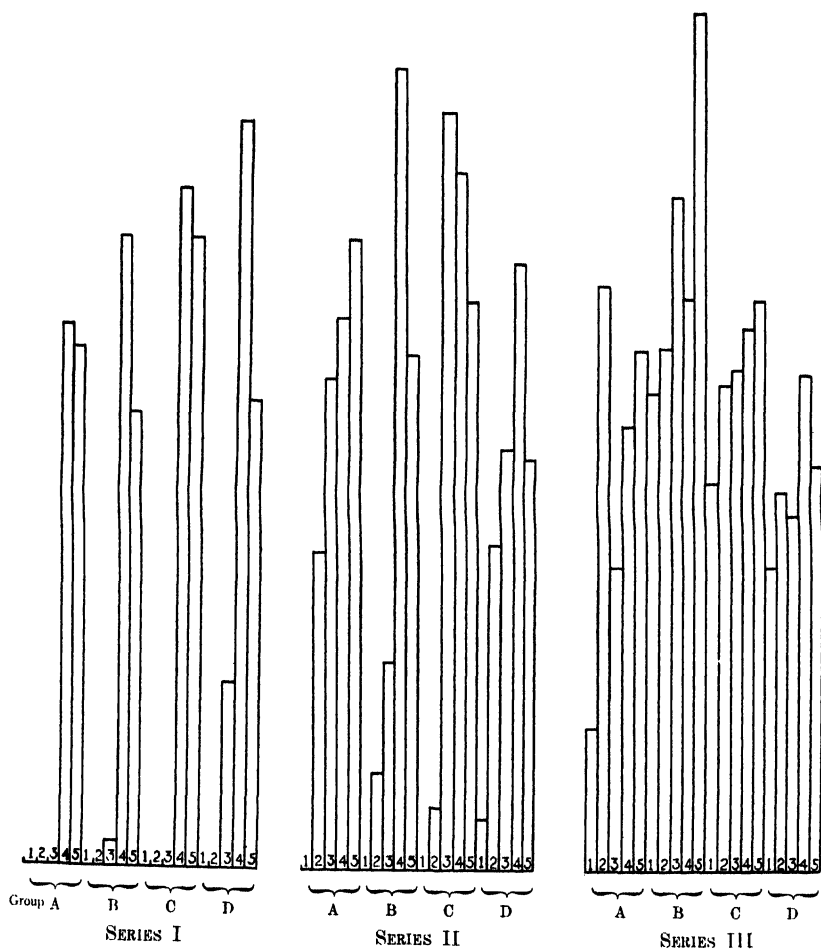


Fig. 4.—Diagrammatic representation of the number of flowers produced by each plant in series I, II, and III.

#### FLOWER PRODUCTION IN SERIES I

The nitrogen supply is seen to have a very definite influence upon flower production. In series I no flowers were produced by plants 1 and 2 of any group and in only two groups were any flowers produced by plant 3, showing definitely that unless nitrogen is present in excess of 0.05 per cent as  $\text{NaNO}_3$ , the undernourished tobacco plant will not flower. Flower production in

general corresponded to vegetative vigor, the plant exhibiting maximum vegetative growth producing the largest number of flowers. Here again the influence of the total concentration of the solution was apparent in the flower yield in the various groups, which increased as the concentration decreased.

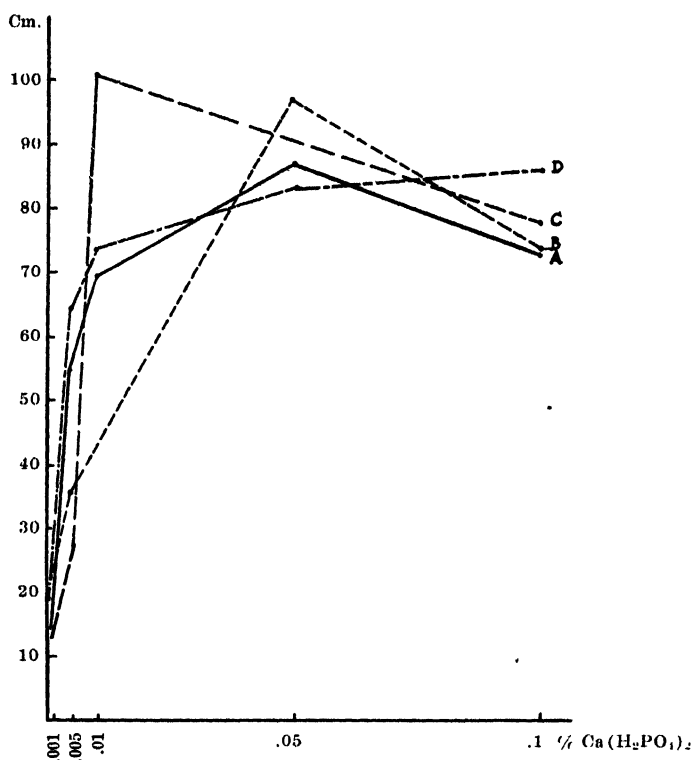


Fig. 5.—Graph showing the influence upon height of equal quantities of  $\text{Ca}(\text{H}_2\text{PO}_4)_2$  in the different groups of series II.

## PHYSIOLOGICAL EFFECT OF PHOSPHORUS

### DISTRIBUTION OF SALTS IN SERIES II

In series II a study was made of the influence of phosphorus added as  $\text{Ca}(\text{H}_2\text{PO}_4)_2$ . This salt was present in the same quantity in all groups A, B, C, and D, the quantity in each of the pots 1 to 5 being 0.02 g. (.001 per cent), 0.1 g. (.005 per cent), 0.2 g. (.01 per cent), 1.0 g. (.05 per cent), 2.0 g. (.1 per cent),

respectively. The maximum weights of the other salts were  $\text{NaNO}_3$ , 3 g.;  $\text{K}_2\text{SO}_4$ , 2.4 g.;  $\text{MgSO}_4$ , 0.96 g., in group A, while the minimum quantities of these salts as added in group D were 0.75 g., 0.6 g., and 0.24 g., respectively. The total concentration in this series ranged from 1.61 g. to 6.38 g.

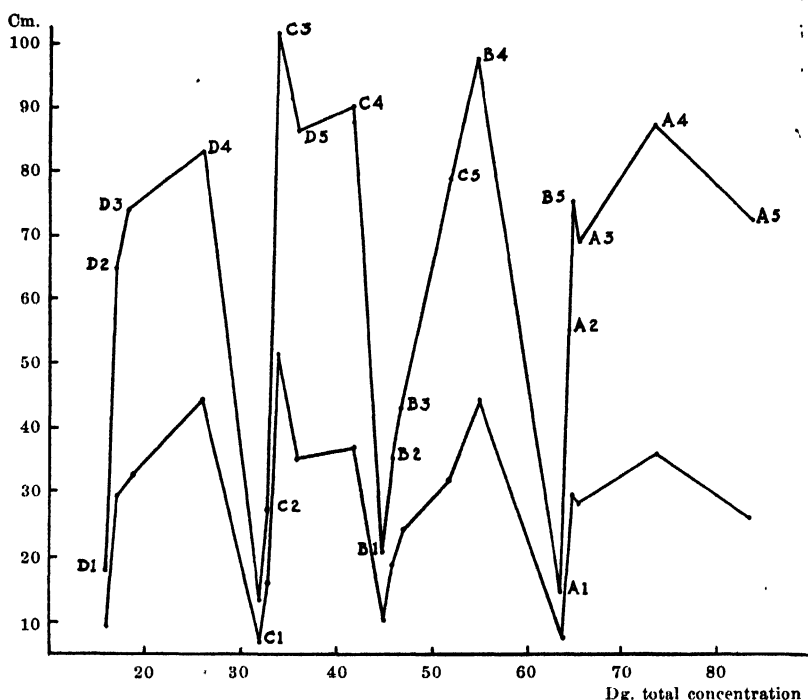


Fig. 6.—Graph showing the influence of total concentration upon height in series II.

#### HEIGHT IN SERIES II

Figure 5 shows the height curves for the five plants of each of the four groups in this series. There is clear indication that the optimum phosphorus supply lies somewhere between .05 per cent and .01 per cent of  $\text{Ca}(\text{H}_2\text{PO}_4)_2$ , the height of plant 4 being greatest when all groups are considered. The difference in height between plants 4 and 5 is not great in any group. The effect of increasing the quantity of  $\text{Ca}(\text{H}_2\text{PO}_4)_2$  is clearly seen in plants 1, 2, 3, and 4 of groups A, B, and D (pl. 8). The effect of total



concentration in this series is not so marked as in series I, but is slightly evidenced in the somewhat better growth of some of the plants in groups C and D as compared with plants in groups A and D (pl. 9). The height of the plants at two different periods in their growth was taken in this series at the same time that the measurements were taken in series I and plotted against total concentration (fig. 6). The total concentration of salts in the solution is here, as in series I, of secondary importance when one element is present in insufficient amount, as will be seen (pl. 6) when plant II C 1 (3.20 g.) is compared with II C 3

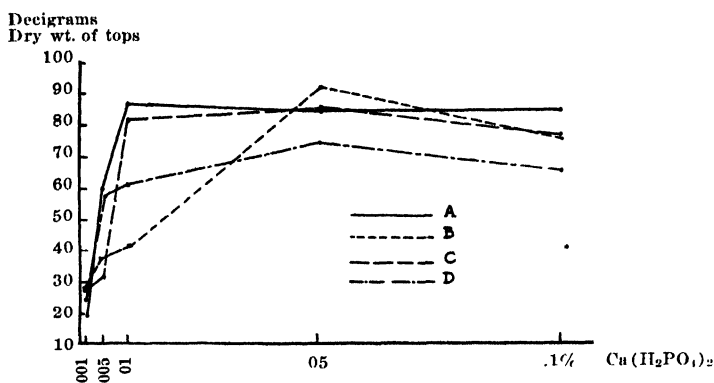


Fig. 7.—Graph showing the influence on dry weight of tops of equal quantities of  $\text{Ca}(\text{H}_2\text{PO}_4)_2$  in the different groups of series II.

(3.38 g.). While the total concentration is practically the same and the total quantity of each of the other salts is the same the  $\text{Ca}(\text{H}_2\text{PO}_4)_2$  in II C 3 is ten times the quantity of the same salt in II C 1.

#### DRY WEIGHT IN SERIES II

A comparison of height (fig. 5) and dry weight curves (fig. 7) for tops in this series shows that height here is not an accurate index to dry weight. Thus group A gave the greatest total dry weight, while the greatest total height of plants occurred in group D, a fact which is explicable since very serious injury resulting in a spindly habit was shown by many plants in the D group, as will be pointed out later in the discussion of injury due to the improper balance of salts in the solution.

### FLOWER PRODUCTION IN SERIES II

The total number of flowers produced in this series was larger than in series I, due largely to the fact that there was less stunted growth in plants 1, 2, and 3 of each group, for which an ample supply of nitrogen was available. Plants II A 1, II B 1, and II C 1 did not produce any flowers. The greatest number of flowers was formed in group C and the greatest yield of any one plant was given by II B 5. The flower yield in each group is definitely related to the general character of the plant as mentioned above in connection with series I.

### PHYSIOLOGICAL EFFECT OF POTASSIUM

#### DISTRIBUTION OF SALTS IN SERIES III

The plants in series III were grown to study the physiological influence of various quantities of potassium added as  $K_2SO_4$  in the presence of a sufficient supply of other nutrient salts, following the same plan as outlined above for the variation of nitrogen and phosphorus in series I and II.  $K_2SO_4$  was present in the same weight in each of the four groups A, B, C, and D, the quantities added to each of the pots 1 to 5 being 0.02 g. (.001 per cent), 0.1 g. (.005 per cent), 0.2 g. (.01 per cent), 1.0 g. (.05 per cent), and 2.0 g. (.1 per cent). The maximum weights of the other salts were  $NaNO_3$ , 3 g.;  $Ca(H_2PO_4)_2$ , 1.2 g.;  $MgSO_4$ , 0.96 g., in group A. In group D the minimum quantities of these salts were 0.75 g., 0.3 g., and 0.24 g., respectively. The total concentration in this series varied from 1.31 g. to 7.16 g.

#### HEIGHT IN SERIES III

No plants in this series were less than 50 cm. high, due to the fact that the two nutrient elements, nitrogen and phosphorus, which in the order named are of first importance as growth factors, were present in sufficient quantity to insure considerable growth (pl. 7). Only a small quantity of  $K_2SO_4$  (.005 to .01 per cent) was required in this series to give plants of maximum height, hence the curves show a downward trend in all plants after either plant 2 or plant 3 in each group (fig. 8, also pl. 12). Thus it is evident that there is a toxic effect of an excess of potassium, irrespective of the total concentration of the solution.

The influence of total concentration is again plainly seen in this series. Plate 13 shows plant 1 of each of the groups A, B, C, and D. As the total concentration decreases from A to C there is a steady increase in the vigor of the plant as judged by height. Curves in figure 9 indicate this general tendency to an increase of height with a decrease in the total concentration of the solution.

### DRY WEIGHT IN SERIES III

As in series I and II, the dry-weight curves (fig. 10) do not correspond with the height curves (fig. 8). The greatest dry weight of any plant was that of III A 4, which was not so tall as the other plants of like number. The low dry weight of III B 4 is especially noticeable. A comparison of the two dry-weight curves for the A and B groups shows a very peculiar effect of  $K_2SO_4$  in solutions of different balance. In each case where the group A curve is high, the group B curve is low, a fact which also applies to plants 3, 4, and 5 in the A and C groups. That this should occur with such regularity is rather remarkable and no satisfactory explanation can as yet be found to account for this situation. It is evident that the physiological balance of salts in the solution is dependent upon the concentration, as has been shown by McCool,<sup>25</sup> Gile,<sup>26</sup> Tottingham,<sup>27</sup> and Shive.<sup>28</sup>

### FLOWER PRODUCTION IN SERIES III

The more vigorous growth of plants in this series gave a greater total yield of flowers than either of the other two series. This was to be expected from the result of series I and II, where the flower yield was shown to be definitely related to the general vigor of the plants. The production of flowers in this series differs, however, from that in the other series. An increase of  $K_2SO_4$ , while in general depressing the total height of the plant, when added in excess of .01 per cent gave a higher flower yield. Thus in group C there is steady increase in the number of flowers produced which corresponds to the increase of  $K_2SO_4$ . The

<sup>25</sup> McCool, Cornell Agric. Exp. Sta. Mem., vol. 2, pp. 121-170, 1913.

<sup>26</sup> Gile, Porto Rico Agric. Exp. Sta. Bull. 12.

<sup>27</sup> *Loc. cit.*

<sup>28</sup> *Loc. cit.*

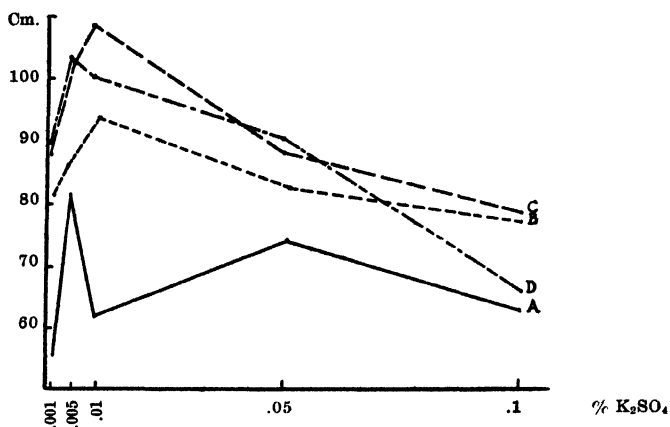


Fig. 8.—Graph showing the influence upon height of equal quantities of  $K_2SO_4$  in the different groups of series III.

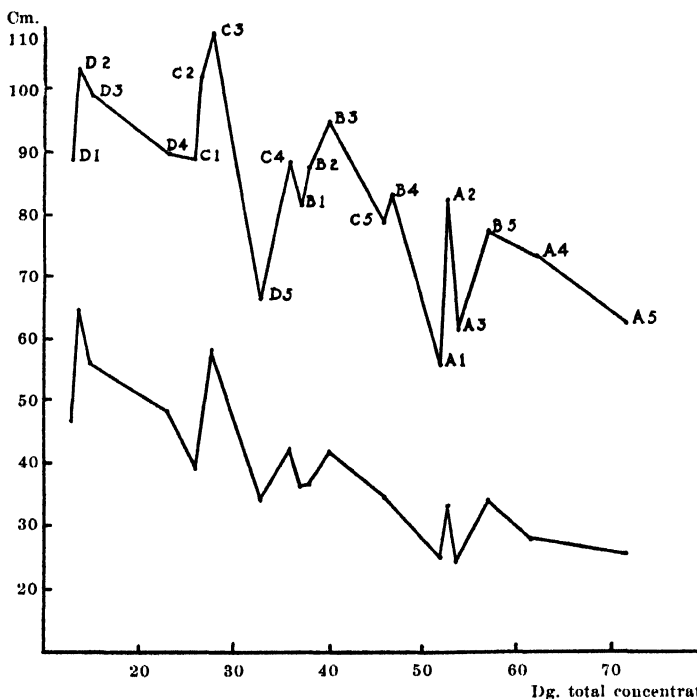


Fig. 9.—Graph showing the influence upon height of total concentration in series III.

greatest flower production occurred in group B, and plant 5 of this group produced a larger number of flowers than any other plant in any series.

#### INJURY DUE TO EXCESS OR DEFICIENCY OF NUTRIENT SALTS

The deficiency or excess of certain elements will cause injury which is very characteristic. Gris as early as 1843 showed that chlorosis was caused by a lack of iron in the nutrient solution. Nitrogen starvation likewise has long been known to cause chlorosis, a condition which may also result from an excess of soluble

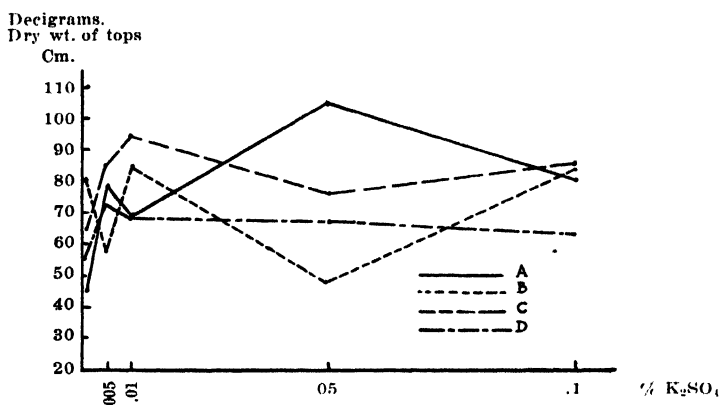


Fig. 10.—Graph showing the influence upon dry weight of tops of equal quantities of  $K_2SO_4$  in the different groups of series III.

phosphate.<sup>29</sup> Magnesium starvation results in injury to the chlorophyll bodies, and in excess is harmful unless antagonized by calcium.<sup>30</sup> Other physiological disturbances not so well established as these mentioned have been considered to be associated with a deficiency or excess of nutrient elements.

The most marked indication of the fact that certain cultures in this experiment did not furnish a normal medium for growth was seen in the chlorosis which is so characteristic of plants grown in solutions which are deficient in nitrogen. This type of injury was uniformly present in all cultures containing 1.0 g. or less of  $NaNO_3$ , but was especially marked in series I, plants 1, 2, and 3, of all groups. It is not possible to draw a sharp

<sup>29</sup> Crone, Sitzungsber. Neiderrhein. Ges. Nat.- und Heilk., Bonn, 1902, pp. 167-173.

<sup>30</sup> Loew and May, U. S. Dept. Agric. Bur. Plant Ind. Bull. 1, 1901.

line which will clearly segregate the injury to the various plants of a large series of this kind into well-defined groups since there is always more or less overlapping. An attempt was made, however, to divide the plants into groups which would show in each case a characteristic type of injury.

(a) This group was made up of plants which were very much stunted, being less than 8 cm. in height and in every case showed a marked chlorosis which was clearly attributed to a low supply of nitrogen. The plants in this group were I A 1, I A 2, I A 3, I B 1, I B 2, I B 3, I C 1, I C 2, I C 3, I D 1.

(b) This group was somewhat taller, from 13 to 21 cm. high, but clearly stunted in growth. These plants showed less chlorosis, since nitrogen was present in sufficient quantity to provide close to the optimum supply in all cultures with the exception of II D 1 and I D 2, where there was a distinct chlorosis due to the low nitrogen supply. Plants showing this type of injury were II A 1, II B 1, II C 1, II D 1, I D 2.

(c) Plants in this group showed a more serious type of injury than any of the other plants. They were more than 40 cm. in height, but were very spindly (pl. 15). The whole plant showed marked chlorosis, which affected the lower leaves most severely and soon resulted in their death. This type of plant was found in cultures I D 3, II B 2, II B 3, II C 2, II D 2, II D 3, II D 4, III D 1, III D 2, III D 3, III D 4, and III D 5.

(d) The plants of this group were decidedly more vigorous than those of the preceding groups, as indicated by their increased height and better color. Indeed, they seemed to be perfectly normal. The following cultures were classed in this group: I C 4, I D 4, II C 3, II C 4, II D 5, III A 1, III A 2, III A 3, III B 1, III B 2, III B 3, III C 1, III C 2, III C 3, III C 4, III C 5. The following plants were even better in appearance than those just named: I A 4, I A 5, I B 4, I B 5, I C 5, I D 5, II A 2, II A 3, II A 4, II A 5, II B 4, II B 5, II C 5, III A 4, III A 5, III B 4, III B 5.

A study of the above grouping indicates that chlorosis was present wherever the nitrogen content was low, as was to be expected. The spindly growth characteristic of groups II D and III D may also be due to the low supply of nitrogen. It seems

that the injury which was so marked in series II must be in some way associated with the calcium-magnesium content in solutions which were not properly balanced. No definite calcium-magnesium ratios can be found which are responsible for the injury which occurred in this series. Twenty different calcium-magnesium ratios ranging from 0.035 to 3.54 occur in the series, and both plants which were normal and plants which were seriously injured were found in cultures in which the calcium-magnesium ratio was low as well as in cultures where this ratio was high. No definite conclusions can be drawn in this connection, however, since the other variables present complicate the situation so that the injury cannot be said with certainty to be associated with an improper balance between the calcium and magnesium.

TABLE 3  
SUMMARY OF COMPLETE EXPERIMENTS

Grouped in such a manner that comparison can be made with special reference to the influence of concentration and balance of the solution upon crop and flower production. The calculation of the per cent total concentration is based on the assumption that the salts are all dissolved in the quantity of water held by 2000 grams of sand when saturated. To get this value in parts per million, multiply the total concentration in grams by 227.2. The real concentration would be much greater than these values would indicate, since the sand was not kept saturated.

SERIES I										
	NaNO <sub>3</sub> , grams	NaNO <sub>3</sub> , per cent	Ca(H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> , grams	K <sub>2</sub> SO <sub>4</sub> , grams	MgSO <sub>4</sub> , grams	Total, grams	Total concentra- tion, per cent in solution	Height, cm.	Tops (dry), grams	Number of flowers
IA1	.02	.001	1.2	2.4	.96	4.58	1.04	2.5	1.12	0.
IB1	.02	.001	.8	1.6	.64	3.06	.69	5.5	1.29	0.
IC1	.02	.001	.6	1.2	.48	2.30	.52	3.5	.74	0.
ID1	.02	.001	.3	.6	.24	1.16	.26	6.5	1.65	0.
IA2	.2	.01	1.2	2.4	.96	4.76	1.08	4.5	1.6	0.
IB2	.2	.01	.8	1.6	.64	3.24	.73	3.5	1.40	0.
IC2	.2	.01	.6	1.2	.48	2.48	.56	2.5	.79	0.
ID2	.2	.01	.3	.6	.24	1.34	.30	17.5	1.53	0.
IA3	1.0	.05	1.2	2.4	.96	5.56	1.26	8.	1.56	0.
IB3	1.0	.05	.8	1.6	.64	4.04	.91	8.	2.17	1.
IC3	1.0	.05	.6	1.2	.48	3.28	.74	4.5	1.65	0.
ID3	1.0	.05	.3	.6	.24	2.14	.48	59.5	5.86	8.5
IA4	2.0	.1	1.2	2.4	.96	6.56	1.35	57.5	.....	25.5
IB4	2.0	.1	.8	1.6	.64	5.04	1.14	78.	8.57	29.
IC4	2.0	.1	.6	1.2	.48	4.28	.97	94.	.....	31.
ID4	2.0	.1	.3	.6	.24	3.14	.71	91.5	11.12	34.5
IA5	3.0	.15	1.2	2.4	.96	7.56	1.71	45.5	7.83	24.
IB5	3.0	.15	.8	1.6	.64	6.04	1.37	49.	7.96	21.
IC5	3.0	.15	.6	1.2	.48	5.28	1.20	74.	9.24	29.
ID5	3.0	.15	.3	.6	.24	4.14	.94	66.	7.60	21.5

## SERIES II

	Ca(H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> grams	Ca(H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> per cent	NaNO <sub>3</sub> grams	K <sub>2</sub> SO <sub>4</sub> grams	MgSO <sub>4</sub> grams	Total, grams	Total concentra- tion, per cent in solution	Height, cm.	Tops (dry)	Number of flowers
IIA1	.02	.001	3.	2.4	.96	6.38	1.45	15.	2.09	0.
IIB1	.02	.001	2.25	1.6	.64	4.51	1.02	21.	2.71	0.
IIC1	.02	.001	1.5	1.2	.48	3.20	.72	13.5	2.73	0.
IID1	.02	.001	.75	.6	.24	1.61	.36	18.	2.47	2.5
IIA2	.10	.005	3.	2.4	.96	6.46	1.46	55.	6.21	14.5
IIB2	.10	.005	2.25	1.6	.64	4.59	1.04	35.5	3.82	4.5
IIC2	.10	.005	1.5	1.2	.48	3.28	.74	27.5	3.22	3.
IID2	.10	.005	.75	.6	.24	1.69	.38	64.5	5.83	15.
IIA3	.20	.01	3.	2.4	.96	6.56	1.49	69.5	8.75	22.5
IIB3	.20	.01	2.25	1.6	.64	4.69	1.06	43.	4.11	9.5
IIC3	.20	.01	1.5	1.2	.48	4.18	.94	101.	8.20	17.5
IID3	.20	.01	.75	.6	.24	2.59	.58	74.	6.27	16.
IIA4	1.0	.05	3.	2.4	.96	7.36	1.67	87.	8.58	25.5
IIB4	1.0	.05	2.25	1.6	.64	5.49	1.24	97.	9.3	37.
IIC4	1.0	.05	1.5	1.2	.48	4.18	.95	90.	8.66	32.
IID4	1.0	.05	.75	.6	.24	2.59	.58	83.5	7.60	28.
IIA5	2.0	.1	3.	2.4	.96	8.36	1.90	73.	8.60	29.
IIB5	2.0	.1	2.25	1.6	.64	6.49	1.47	74.5	7.76	23.5
IIC5	2.0	.1	1.5	1.2	.48	5.18	1.17	78.	7.88	26.5
IID5	2.0	.1	.75	.6	.24	3.59	.81	86.	6.75	19.

## SERIES III

	K <sub>2</sub> SO <sub>4</sub> grams	K <sub>2</sub> SO <sub>4</sub> per cent	NaNO <sub>3</sub> grams	Ca(H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> grams	MgSO <sub>4</sub> grams	Total, grams	Total concentra- tion, per cent in solution	Height, cm.	Tops (dry)	Number of flowers
IIIA1	.02	.001	3.	1.2	.96	5.18	1.17	56.5	4.65	6.5
IIIB1	.02	.001	2.25	.8	.64	3.71	.84	81.5	8.18	22.
IIIC1	.02	.001	1.5	.6	.48	2.60	.59	89.	6.58	18.
IID1	.02	.001	.75	.3	.24	1.31	.29	89.	5.56	14.
IIIA2	.1	.005	3.	1.2	.96	5.26	1.19	82.5	7.88	27.
IIIB2	.1	.005	2.25	.8	.64	3.79	.86	87.	5.87	24.
IIIC2	.1	.005	1.5	.6	.48	2.68	.60	102.	8.57	22.5
IID2	.1	.005	.75	.3	.24	1.39	.31	103.5	7.27	17.5
IIIA3	.2	.01	3.	1.2	.96	5.36	1.21	62.	6.97	14.
IIIB3	.2	.01	2.25	.8	.64	3.89	.88	94.5	8.4	31.
IIIC3	.2	.01	1.5	.6	.48	2.78	.63	109.	9.4	23.
IID3	.2	.01	.75	.3	.24	1.49	.33	100.5	6.9	16.5
IIIA4	1.0	.05	3.	1.2	.96	6.16	1.40	74.	10.53	20.5
IIIB4	1.0	.05	2.25	.8	.64	4.69	1.06	83.	4.86	26.5
IIIC4	1.0	.05	1.5	.6	.48	3.58	.81	88.5	7.66	25.
IID4	1.0	.05	.75	.3	.24	2.29	.52	90.	6.75	23.
IIIA5	2.0	.1	3.	1.2	.96	7.16	1.62	63.	8.11	24.
IIIB5	2.0	.1	2.25	.8	.64	5.69	1.29	77.5	8.42	39.5
IIIC5	2.0	.1	1.5	.6	.48	4.58	1.04	79.	8.68	26.5
IID5	2.0	.1	.75	.3	.24	3.29	.74	66.5	6.36	19.



## THE ABSORPTION OF SALTS FROM THE CULTURE MEDIUM

The absorption by plants of salts from a solution is no doubt intimately related to the composition of the solution, since the intake of inorganic salts occurs only when the concentration of the solute outside the permeable protoplast is greater than that within. Hence the total osmotic concentration of a solution may affect the intake and storage of the salts from the solution.<sup>31</sup> The quantity of salts which are absorbed by the plant may also vary with the qualitative composition of the solution. Thus True and Bartlett<sup>32</sup> have shown that absorption from a solution of two or of three salts is more rapid than from a solution containing a single salt. It is evident that the factors which regulate absorption are very complex, and that this complexity increases with an increase in the number of different anions and cations in the solution. It is further evident that two methods may be followed with a view to ascertaining the quantities of salts absorbed. The ash of the plants may be analyzed or the residue of salts remaining in the culture may be determined. It has seemed desirable to attempt a study of absorption in the present case by means of the latter method.

## METHOD OF ANALYSIS

The small quantities of salts added to each pot made the acid-extraction method undesirable, since the large quantities of sand required to give weighable precipitates would have been extremely hard to dehydrate, thus introducing a considerable source of error. For this reason the colorimetric method of analysis<sup>33</sup> was used in this work. An exception was made in the determination of calcium, which was made by the usual volumetric method. The sulphur was determined gravimetrically as  $\text{BaSO}_4$ . A water extract was prepared from 250 g. of sand by leaching with successive small portions of distilled water until

<sup>31</sup> Livingston, *The rôle of diffusion and osmotic pressure in plants*, Chicago, 1903.

<sup>32</sup> True and Bartlett, *U. S. Dept. Agric. Bur. Plant Ind. Bull.* 231, 1912, and *Am. Jour. Bot.*, vol. 2, pp. 255-278, 311-323, 1915, vol. 3, pp. 47-58, 1916.

<sup>33</sup> Schreiner and Failyer, *U. S. Dept. Agric. Bur. Soils, Bull.* 31, 1906.

500 c.c. had been used. The filter containing the sand was then allowed to stand for about fifteen hours, and the sand was leached with another 500 c.c. portion of water. The filtrate was evaporated to dryness, the residue dehydrated at 100° C, and strongly ignited in a platinum dish to destroy organic matter. The residue was then dissolved in hot water and made up to definite volume from which aliquot portions were taken for analysis. A 2000 g. sample of the washed sand used in the experiment gave by this method the following analysis:

N	K	P	Mg	Ca	S
00.	33.6	5.2	66.4	2.4	15.6 milligrams

In this analysis, as well as in all the following analyses, the statement made is for the total weight in milligrams of the element contained in 2000 g. of sand, which was the quantity contained in each pot. Table 4 gives a summary of the analysis of each of the sixty samples of sand used. The total quantity of each element present, calculated from the quantities of salts added plus the quantity in the original sand, is given, as well as the number of milligrams of the element missing from the pot at the end of the growing season. This latter value is the difference between the total weight of the elements present and the residue as shown by the analysis of the water extract. The difference thus obtained represents the quantity of the element absorbed by the plant and adsorbed by the sand particles. It must be admitted that it is not known just how great a factor adsorption may be in the case of this sand.

#### DISCUSSION OF RESULTS

In series I no nitrogen as nitrate remained in pots 1, 2, 3, and 4 of groups A, B, and C. In group D there is a small residue of nitrogen in spite of the fact that the total crop production was greatest in this group. The concentration of the solution clearly affects the economical use of nitrogen, a fact which in general is also indicated in series II and in series III. It is especially noticeable that there is a much larger quantity of nitrogen left in series III D than in either series I D or II D. The total crop production for group III D was better than that of the D group



	N total	N remaining	N difference	P total	P remaining	P difference	K total	K remaining	K difference	Mg total	Mg remaining	Mg difference	Ca total	Ca remaining	Ca difference	Qa total	Qa remaining	Qa difference	Dry weight, grams
IIB1	370.4	1.26	369.1	10.1	2.9	7.2	392.8	68.8	324.0	129.4	71.2	58.2	5.6	20.8	.....	391.3	36.4	354.9	2.71
IIB2	370.4	1.08	369.3	29.8	2.3	27.5	392.8	57.6	335.2	129.4	45.6	83.8	18.3	52.8	.....	391.3	36.8	354.5	3.82
IIB3	370.4	.81	369.5	54.8	3.1	51.2	392.8	60.8	332.0	129.4	64.0	65.4	34.1	20.8	8.3	391.3	33.4	357.9	4.11
IIB4	370.4	.90	369.5	251.1	7.5	243.6	392.8	67.2	325.6	129.4	62.8	66.6	161.0	44.8	116.2	391.3	28.0	363.3	9.30
IIB5	370.4	1.35	369.0	497.1	8.6	488.5	392.8	86.4	306.4	129.4	64.0	65.4	319.7	41.6	278.1	391.3	29.6	361.7	7.76
IIIC1	246.9	.63	246.3	10.1	3.1	7.0	303.0	72.0	231.0	113.7	48.4	58.2	5.6	44.8	.....	297.2	32.9	264.3	2.73
IIIC2	246.9	.90	246.0	29.8	2.3	27.5	303.0	70.4	232.6	113.7	56.8	56.9	18.3	70.4	.....	297.2	27.4	269.8	3.22
IIIC3	246.9	.....	.....	54.3	3.4	50.9	303.0	52.8	250.2	113.7	71.2	42.5	34.1	13.6	20.5	297.2	51.6	245.6	8.20
IIIC4	246.9	.54	246.3	251.1	6.5	244.6	303.0	46.4	256.6	113.7	56.8	56.9	161.0	57.6	103.4	297.2	42.2	255.0	8.66
IIIC5	246.9	1.39	245.5	497.1	7.5	489.6	303.0	83.6	219.4	113.7	74.0	39.7	319.7	24.8	294.9	297.2	19.7	277.5	7.88
IID1	123.5	.45	123.0	10.1	2.3	7.8	168.3	56.0	112.3	90.0	51.2	38.8	5.6	24.0	.....	156.2	19.4	136.8	2.47
IID2	123.5	.81	122.7	29.8	2.6	27.2	168.3	32.0	136.3	90.0	56.8	33.2	18.3	12.8	5.5	156.2	19.4	136.8	5.83
IID3	123.5	.76	122.7	54.3	3.4	50.9	168.3	20.8	147.5	90.0	60.8	29.2	34.1	9.6	24.5	156.2	27.4	128.8	6.27
IID4	123.5	.45	122.0	251.1	3.6	247.5	168.3	38.4	129.9	90.0	78.0	12.0	161.0	8.0	153.0	156.2	20.9	135.4	7.60
IID5	123.5	.45	122.0	497.1	5.8	491.3	168.3	37.6	130.7	90.0	79.6	10.4	319.7	17.6	302.1	156.2	8.2	148.0	6.75
IIIA1	493.9	1.8	492.1	300.3	4.6	295.7	38.1	16.4	21.7	160.9	49.6	111.3	192.8	12.0	180.8	143.0	10.9	132.1	4.65
IIIA2	493.9	3.6	490.3	300.3	4.6	295.7	56.0	28.0	28.0	160.9	74.0	86.9	192.8	7.2	185.6	158.2	33.4	124.8	7.88
IIIA3	493.9	2.7	491.2	300.3	6.5	293.8	78.6	22.0	56.6	160.9	61.2	99.7	192.8	19.2	173.6	171.5	24.6	146.9	6.97
IIIA4	493.9	2.3	491.6	300.3	6.0	294.3	258.0	57.6	200.4	160.9	.....	.....	.....	.....	.....	322.9	60.4	262.5	10.53
IIIA5	493.9	2.3	491.6	300.3	7.3	293.0	483.6	29.6	454.0	160.9	71.2	89.7	192.8	17.6	175.2	455.9	.....	.....	8.11
IIIB1	370.4	3.61	366.8	207.9	6.0	195.9	38.1	26.4	11.7	129.4	35.6	93.8	129.3	10.4	118.9	101.6	15.6	86.0	8.18
IIIB2	370.4	2.25	368.1	207.9	6.2	195.7	56.0	40.0	16.0	129.4	68.4	61.0	129.3	23.2	106.1	1.68	83.9	77.9	5.87
IIIB3	370.4	1.35	369.0	207.9	5.8	196.1	78.6	24.0	54.6	129.4	49.6	79.8	129.3	10.4	118.9	129.1	36.9	92.2	8.84
IIIB4	370.4	1.44	368.9	207.9	5.8	196.1	258.0	43.2	214.8	129.4	99.6	29.8	129.3	4.8	124.5	281.5	10.2	271.3	4.86
IIIB5	370.4	.85	369.5	207.9	5.0	196.9	483.6	67.2	416.4	129.4	96.8	32.6	129.3	8.0	131.3	414.5	59.8	344.7	8.82
IIIC1	246.9	.36	246.5	152.8	2.9	149.9	38.1	28.0	10.1	113.7	82.8	30.9	97.6	8.0	89.6	80.7	17.5	63.2	6.58
IIIC2	246.9	1.58	245.3	152.8	4.4	148.4	56.0	18.4	37.6	113.7	75.2	38.5	97.6	8.8	88.8	95.9	.....	.....	8.57
IIIC3	246.9	.77	246.1	152.8	3.8	149.0	78.6	35.2	43.4	113.7	91.2	22.5	97.6	7.2	90.4	109.2	28.5	81.7	9.40
IIIC4	246.9	.41	246.5	152.8	7.0	145.8	258.0	41.6	216.4	113.7	62.8	50.9	97.6	10.4	87.2	260.6	50.0	210.1	7.66
IIIC5	246.9	4.52	242.4	152.8	7.3	145.5	483.6	60.8	422.8	113.7	68.4	45.3	97.6	11.2	86.4	393.6	30.7	262.9	8.68
IIID1	123.5	4.52	118.9	79.0	5.8	73.2	38.1	18.4	19.7	90.0	48.4	41.6	50.0	7.2	42.8	49.5	28.0	21.5	5.56
IIID2	123.5	4.06	119.4	79.0	4.1	74.9	56.0	18.4	37.6	90.0	85.6	4.4	50.0	6.4	43.6	54.7	23.0	31.7	7.29
IIID3	123.5	3.16	120.3	79.0	6.0	73.0	78.6	17.6	61.0	90.0	68.4	21.4	50.0	9.6	40.4	78.0	19.7	58.3	6.96
IIID4	123.5	4.52	118.9	79.0	6.5	72.5	258.0	40.0	218.0	90.0	89.2	.....	50.0	7.2	42.8	239.4	29.1	200.3	6.75
IIID5	123.5	4.96	118.5	79.0	6.0	73.0	483.6	48.0	435.6	90.0	84.0	6.0	50.0	3.2	46.8	362.4	45.0	317.4	6.36

in either of the other series, a result which is explicable since nitrogen is a more important limiting factor than phosphorus, and phosphorus is in turn more important as a limiting factor in growth than potassium. Hence in a solution where nitrogen is deficient, and potassium and phosphorus present in optimum amounts, a smaller crop production results than is the case where phosphorus is the deficient factor. A deficiency of potassium does not so seriously affect the intake of other salts, with the result that a better crop is produced than is the case where phosphorus is the deficient factor.

The method of analysis which it was necessary to use makes the analytical values for phosphorus and magnesium rather unreliable. In series II, where phosphate was added in increasing quantities from pot 1 to pot 5 in each group, the analysis of the water extract shows the effect of this increment. It will be noticed that the analytical values are all of about the same order of magnitude, which may point strongly to adsorption by the sand. No general conclusion as to the adsorption of phosphorus and of magnesium can be drawn for the reasons above enumerated.

In the case of potassium the method of analysis was much more accurate. There is evidence in series I, groups A, B, C, and D, of an increase in the amount of potassium absorbed which in general seems to be related to the more vigorous growth resulting from the increasing quantities of nitrogen as added in these groups. A similar relation holds in the other two series, as will be seen from the ratios between potassium added and potassium remaining, which are higher where the nitrogen is added in large quantities than is the case where this element was present in small quantities. The high ratios of potassium added to potassium remaining after growth, therefore, usually occur where the dry weight is highest.

Not much can be said on the absorption of calcium. In series I it is noticeable that the quantity of calcium remaining, as compared in the four groups of this series, bears an inverse relation to the calcium added. Less calcium is absorbed from the solutions of low total concentration than from those which have a high concentration. Since the crop production was

greater in group D, the calcium must have been used with greater economy in this group where the total concentration was low.

The large number of variables which are present in each of the solutions make accurate deductions concerning the exact relation between any element and the growth of the plant almost impossible. In fact, growth has been shown to be influenced not by one factor alone but by combinations of factors.

### GENERAL DISCUSSION

It is evident, as noted above, that the large number of variables present in an experiment of this character so complicates the situation that definite conclusions are drawn only with considerable difficulty. Inorganic salts can be used by the plant only from solution. The complexity of this solution increases with the number of ions, which must be rather large since the plant cannot make normal growth unless certain ions are present. To further complicate the situation, all the salts may be available which are required for growth, but the unbalanced condition of the solution may cause injury to the growing plant.<sup>34</sup> This condition of balance, in turn, seems to be related to the total concentration of the solution as well as to its qualitative composition.<sup>35</sup> In this connection it is important to note that if it were possible to keep the balance in the solution constant by renewal of salts, growth differences would be less marked than when the plant grows in a solution in which the balance is constantly changing due to absorption of ions by the plant.<sup>36</sup> All of the above points must be taken into consideration in any experimental work which is done in this field of investigation. The exact influence which the concentration of the solution has upon the complicated physiological processes concerned in plant nutrition is a problem which can be solved only by the gradual accumulation of a mass of evidence bearing upon the subject. The complexity of the whole problem is such as to require more than

<sup>34</sup> Loeb, *Archiv. ges. Physiol.*, vol. 88, pp. 68-78, 1902, and *Amer. Jour. Physiol.*, vol. 3, pp. 327-338, 1900; Osterhout, *Science*, n. s., vol. 35, pp. 112-115, 1912, and *Jour. Biol. Chem.*, vol. 1, p. 363, 1906.

<sup>35</sup> Gile, *loc. cit.*

<sup>36</sup> Brenchley, *Ann. Bot.*, vol. 30, pp. 77-90, 1916.

the evidence of a single set of experiments for proof. The evidence presented by the work herein reported is an addition to that already reported by other investigators, who have shown that the absorption is influenced to a more or less marked degree by the concentration of the solution. Conclusions which have been reached in regard to the effect of certain variables in this study must be understood to apply only in the case of the specific combinations of salts studied, and in connection with the growth of the tobacco plant in sand cultures.

The experiments herein reported have in part been made possible by that portion of the Adams fund allotment of the Department of Agriculture of the University of California placed at the disposal of Professor W. A. Setchell of the Department of Botany. It is a pleasure to acknowledge indebtedness to Professor Charles B. Lipman and to Dr. T. H. Goodspeed, who have by helpful advice and criticism directed the work.

## SUMMARY

Results are above given which deal with the influence of the composition and concentration of the nutrient solution on sixty different plants of an  $F_1$  species-hybrid of *Nicotiana*.

1. 2000 g. of washed sand of known composition was used as a culture medium for each plant.

2. The salts used were  $\text{NaNO}_3$ ,  $\text{Ca}(\text{H}_2\text{PO}_4)_2$ ,  $\text{K}_2\text{SO}_4$ , and  $\text{MgSO}_4$ .

3. The salts were so distributed as to give at least three important variables: first, as to a single nutrient salt; second, as to total concentration of salts; and third, as to the balance of salts in the solution.

4. The influence of the solution on the growth of the plant was judged by the following criteria: height, leaf length, leaf width, flower production, dry weight of tops.

5. Nitrogen is a more important growth-limiting factor than phosphorus, and phosphorus is, in turn, more important in this capacity than potassium.

6. The total concentration of the solution has a marked influence upon growth. Plants growing in solutions of low concen-

tration were in general superior to those grown in solutions of higher concentration.

7. Flower yield as well as vegetative vigor is influenced by the composition and concentration of the nutrient solution.

8. The physiological balance of salts in the solution is an important factor which must be taken into consideration in connection with the composition and concentration of the solution. Growth is influenced by a combination of all of these factors.

9. A quantitative analysis of the sand used in each pot was made after the plants were harvested.

10. Evidence of adsorption is seen in the results of the quantitative analysis of the water extract from the sand.

11. The concentration of the nutrient solution clearly affects the economical use of nitrogen.

12. High ratios of potassium added to potassium remaining after growth usually occur where the dry weight production is greatest.

13. Less calcium is absorbed from solutions of low total concentration than from those which have a high total concentration.

14. Calcium seems to be used with greater economy in solutions where the total concentration is low than in solutions in which the total concentration is high.

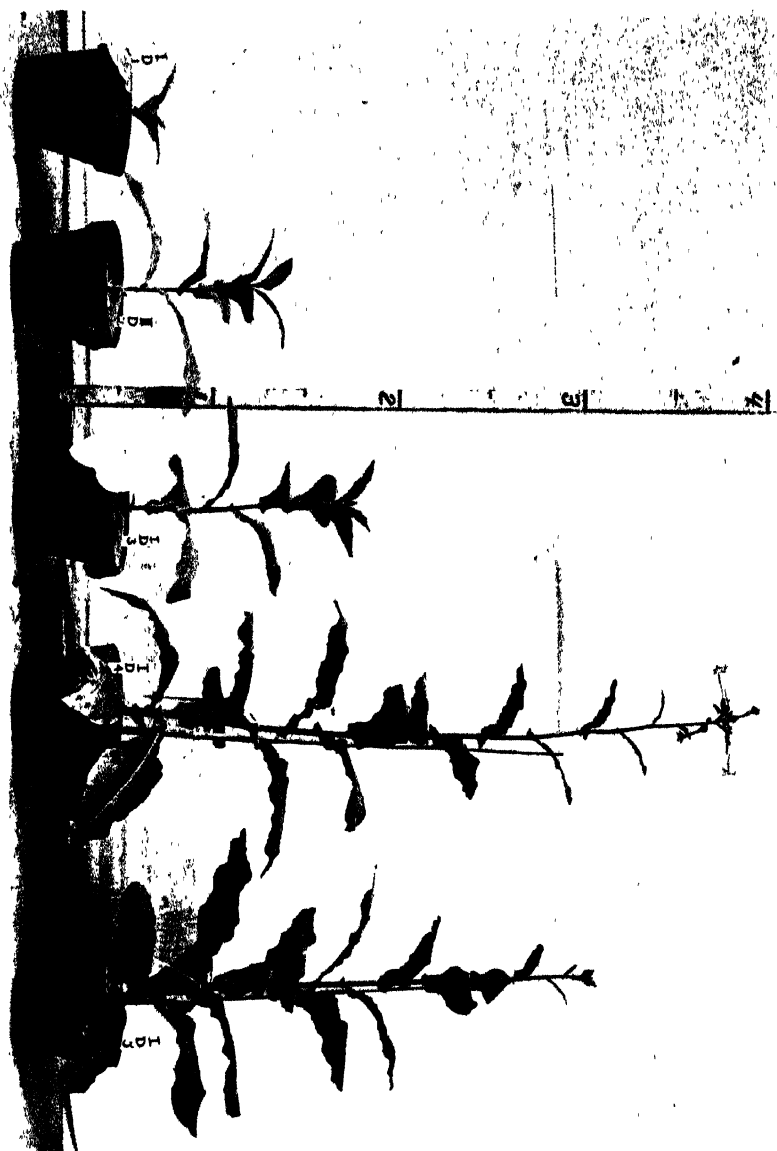
*Transmitted April 21, 1916.*



# PLATE 5

## TREATMENT

	NaNO <sub>3</sub> Grams	Ca(H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> Grams	Total Grams
Pot 1D1	.02	.3	.6	.24	1.16
Pot 1D2	.2	.3	.6	.24	1.34
Pot 1D3	1.0	.3	.6	.24	2.14
Pot 1D4	2.0	.3	.6	.24	3.14
Pot 1D5	3.0	.3	.6	.24	4.14



# PLATE 6

## TREATMENT

	NaNO <sub>3</sub> Grams	Ca (H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> Grams	Total Grams
Pot IA4	2.0	1.2	2.4	.96	6.56
Pot IB4	2.0	.8	1.6	.64	5.04
Pot IC4	2.0	.6	1.2	.48	4.28
Pot ID4	2.0	.3	.6	.24	3.14



# PLATE 7

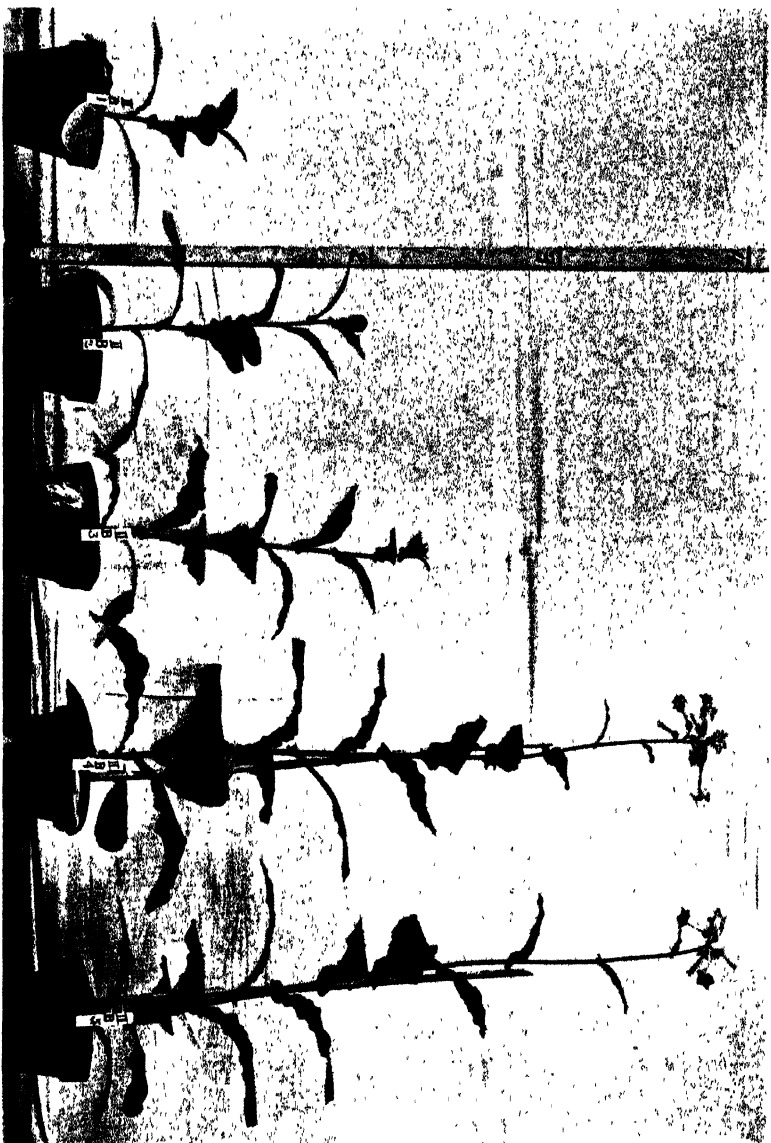
## TREATMENT

	NaNO <sub>3</sub> Grams	Ca (H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> Grams	Total Grams
Pot IC1	.02	.6	1.2	.48	2.30
Pot ID3	1.0	.3	.6	.24	2.14
Pot IB1	.02	.8	1.6	.64	3.06
Pot ID4	2.0	.3	.6	.24	3.14



# PLATE 8

		TREATMENT				
		NaNO <sub>3</sub> Grams	Ca (H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> Grams	Total Grams
Pot	IIB1	2.25	.02	1.6	.64	4.51
Pot	IIB2	2.25	.1	1.6	.64	4.59
Pot	IIB3	2.25	.2	1.6	.64	4.69
Pot	IIB4	2.25	1.0	1.6	.64	5.49
Pot	IIB5	2.25	2.0	1.6	.64	6.49





# PLATE 9

## TREATMENT

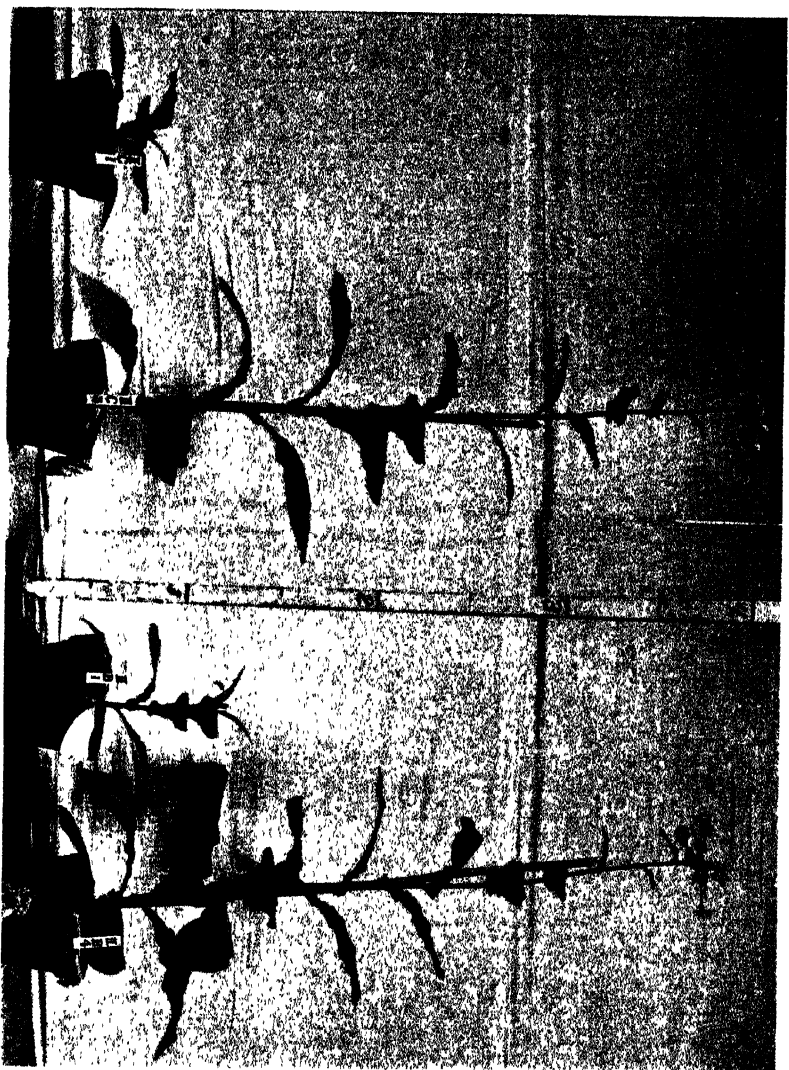
	NaNO <sub>3</sub> Grams	Ca (H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> Grams	Total Grams
Pot IIA3	3.00	.2	2.4	.96	6.56
Pot IIB3	2.25	.2	1.6	.64	4.69
Pot IIC3	1.5	.2	1.2	.48	3.38
Pot IID3	.75	.2	.6	.24	1.79



# PLATE 10

## TREATMENT

	NaNO <sub>3</sub> Grams	Ca(H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> Grams	Total Grams
Pot IIC1	1.5	.02	1.2	.48	3.20
Pot IIC3	1.5	.2	1.2	.48	3.38
Pot IIB1	2.25	.02	1.6	.64	4.51
Pot IIB4	2.25	1.0	1.6	.64	5.49



# PLATE 11

## TREATMENT

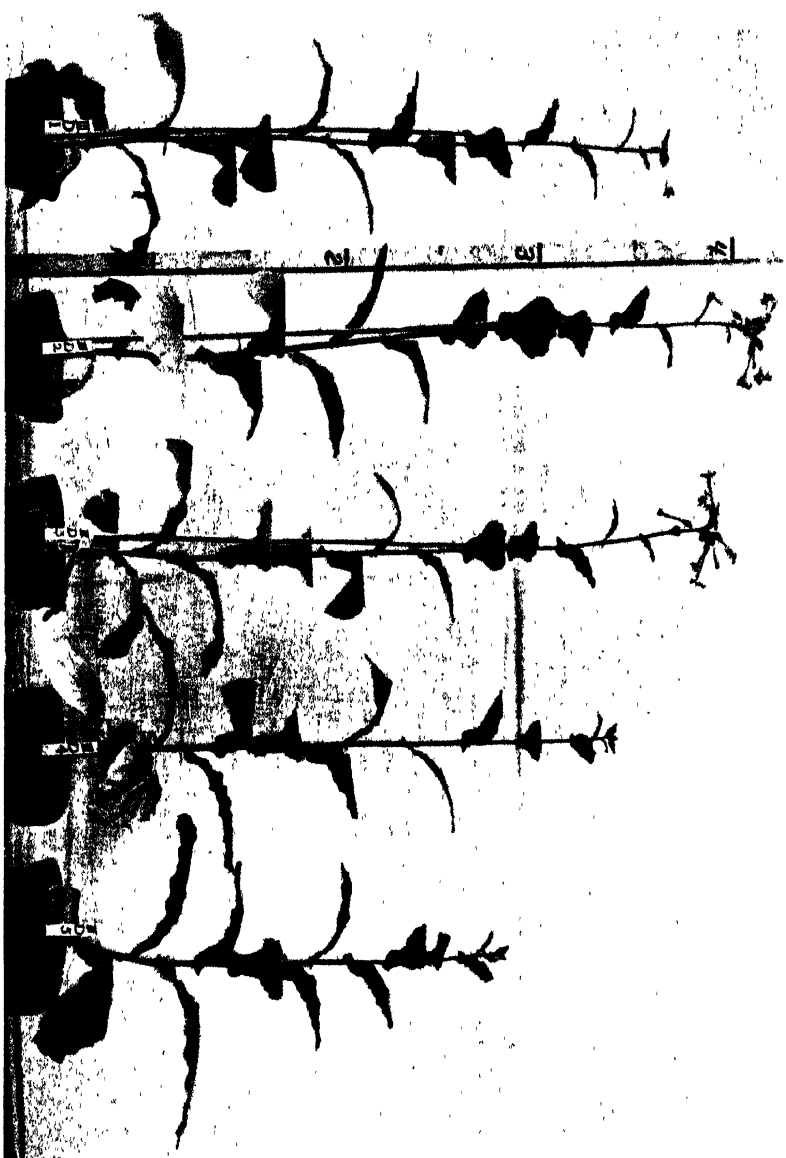
	NaNO <sub>3</sub> Grams	Ca (H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> Grams	Total Grams
Pot 1B3	1.00	.8	1.6	.64	4.04
Pot IIB3	2.25	.2	1.6	.64	4.69
Pot IIIB3	2.25	.8	.2	.64	3.89



# PLATE 12

## TREATMENT

	NaNO <sub>3</sub> Grams	Ca (H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> Grams	Total Grams
Pot 111D1	.75	.3	.02	.24	1.31
Pot 111D2	.75	.3	.1	.24	1.39
Pot 111D3	.75	.3	.2	.24	1.49
Pot 111D4	.75	.3	1.0	.24	2.29
Pot 111D5	.75	.3	2.0	.24	3.29





# PLATE 13

## TREATMENT

	NaNO <sub>3</sub> Grams	Ca(H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> Grams	Total Grams
Pot IIA1	3.00	1.2	.02	.96	5.18
Pot IIIB1	2.25	.8	.02	.64	3.71
Pot IIIC1	1.5	.6	.02	.48	2.60
Pot IIID1	.75	.3	.02	.24	1.31



# PLATE 14

## TREATMENT

Plant growing in washed sand

Plant growing in soil

	NaNO <sub>3</sub> Grams	Ca (H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> · Grams	Total Grams
Pot 111B2	2.25	.8	.1	.64	3.79



# PLATE 15

## TREATMENT

	NaNO <sub>3</sub> Grams	Ca (H <sub>2</sub> PO <sub>4</sub> ) <sub>2</sub> Grams	K <sub>2</sub> SO <sub>4</sub> Grams	MgSO <sub>4</sub> Grams	Total Grams
Pot ID2	.2	.3	.6	.24	1.34
Pot IID2	.75	.1	.6	.24	1.69
Pot IIID2	.75	.3	.1	.24	1.39



**UNIVERSITY OF CALIFORNIA PUBLICATIONS**  
**IN**  
**AGRICULTURAL SCIENCES**

Vol. 1, No. 12, pp. 395-494, plates 6-9

April 7, 1917

**CERTAIN EFFECTS UNDER IRRIGATION**  
**OF COPPER COMPOUNDS UPON CROPS**

**BY**  
**R. H. FORBES**

**UNIVERSITY OF CALIFORNIA PRESS**  
**BERKELEY**

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## CERTAIN EFFECTS UNDER IRRIGATION OF COPPER COMPOUNDS UPON CROPS\*

By R. H. FORBES

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\* Paper No. 31, Citrus Experiment Station, College of Agriculture, University of California, Riverside, California.

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## Part I.—EXPERIMENTAL WORK

### INTRODUCTION

The region to which the studies described in this publication more particularly relate lies in southeastern Arizona in Greenlee and Graham counties and consists, first, of the Clifton-Morenci mining district and second, of the irrigated lands along the Gila River from twenty-five to sixty miles below. The Clifton-Morenci mining district is drained by Chase Creek into the San Francisco River, which in turn empties into the Gila. From the Gila, beginning at a point about twenty-five miles by channel below Clifton, irrigating waters are withdrawn for the use of the rich lands extending somewhat discontinuously from above San Jose to Fort Thomas, a distance of thirty miles. For about forty years, this up-stream mining district and the irrigated lands below have developed together from small beginnings into large industries.

Beginning with the initiation of smelting operations on the

San Francisco River in 1882, comparatively small amounts of mining detritus must have found their way into the irrigating water-supply. Following the discovery, in 1893, of immense deposits of low-grade sulphide ores in the district and the erection of concentrating plants to handle them, rapidly increasing quantities of fine slimes were discharged into the stream-flow, becoming noticeable in the irrigating waters of Graham County about the year 1900. Following the observation of their presence, various crop failures were attributed from time to time to the tailings, resulting finally in a request by the farmers of the district to the writer, for an examination of the facts relating to damage done by mining detritus to their irrigated crops.

#### SOLID WASTES

Following this request, the writer began a study of the problem in May, 1904, which resulted in the publication of Bulletin 53 of the Arizona Agricultural Experiment Station, September 20, 1906. This publication established the fact that irrigating sediments, in general, may be beneficial or harmful according to their composition and physical character and to the manner of their disposition in or upon the soil. If allowed to accumulate upon the surface of the soil in the form of more or less impervious silt-blankets, their influence, by limiting the supply of water and air to the soil, is notably harmful. In the case of the mining wastes from the Clifton-Morenci district, which are particularly plastic and "tight" in character, the damage done was found to be greater than that resulting from sediments arising from ordinary erosion. It was determined that the damage from these wastes, particularly to alfalfa and other crops which cannot receive constant and thorough cultivation, was of an increasingly serious character.

The farmers of Graham County, represented by one of their number, finally brought suit against the Arizona Copper Company, Limited, for discharging tailings into their irrigating water-supply. The case was decided in the District Court of Graham County in favor of the farmers, and an order was issued in November, 1907, effective May 1, 1908, restraining the mining companies from discharging "slimes, slickens or tailings" into

Chase Creek, the San Francisco River, or the Gila River. The case was appealed to the territorial Supreme Court where, however, the decision was confirmed in March, 1909. The case was again appealed by the Arizona Copper Company to the Supreme

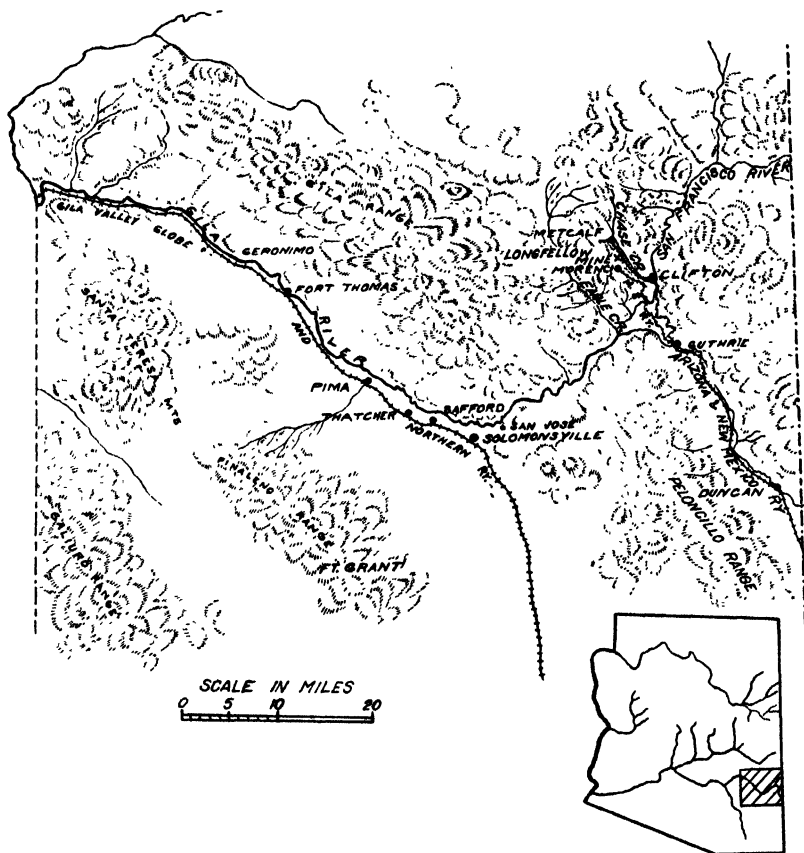


Fig. 1.—General map of the Clifton-Morenci and Gila River mining and irrigation district, Arizona

Court of the United States, where it was again and finally decided in favor of the farmers on June 16, 1913.

During and since the occurrences above mentioned, large quantities of solid wastes have been impounded by the copper companies in settling basins constructed for their storage in the

district. Recent investigations by the companies indicate a possibility that with copper at 15 cents a pound these stored tailings, which average about 0.85 per cent copper, may be profitably reworked.

In the long run, therefore, it may be found that an adjustment based upon a complete and impartial statement of facts relating to the tailings situation is beneficial both to the agricultural and to the mining interests concerned.

#### SOLUBLE COPPER COMPOUNDS

Following the disposition of mining detritus, there remained the problem of soluble copper compounds which, in small but continuously appreciable quantities, find their way with waste waters into the stream-flow of the region. These compounds originate in the ores of the district and are, as in the case of the carbonates, directly soluble to a slight extent in drainage waters, especially in the presence of carbon dioxide. In other cases, the original ores are changed through the action of air into soluble substances which then escape downstream. Sulphide ores are thus oxidized in the presence of air into soluble copper sulphate. Inasmuch as it is well known that minute amounts of copper in solution are extremely toxic to plant roots directly exposed to them, some apprehension naturally existed as to the effects of these small amounts of copper salts escaping into the water-supply of an irrigated district.

In some respects, conditions were especially favorable here to the successful prosecution of a study of the foregoing question. The irrigated lands are at a distance of twenty miles or more from the smelters, so that injurious gases could not complicate effects upon irrigated crops. There are, also, only traces of other toxic metals to be found within the district—more particularly, arsenic, antimony, and zinc. Injurious effects due to the possible toxic action of compounds originating in the mines are therefore limited to copper.

Scientific study relating to toxic effects of copper upon plants under varying conditions has thoroughly established not only the fact that copper compounds are extremely toxic to plants when they obtain entry to their tissues, but also that various

agencies standing between these poisonous salts and the living plant tend to prevent injury.<sup>1</sup> Soluble copper compounds, for instance, react with carbonate of lime, commonly abundant in soils of the arid region, to form the solid carbonates of copper. The partly decomposed silicates of these soils also precipitate soluble compounds of copper and mask their toxic character. Organic matter in the soil likewise holds large quantities of copper in comparatively harmless combinations. Through physical attraction or *adsorption*, soluble copper compounds enter into weak combination with fine soil particles and toxic effects are thereby greatly lessened. In the presence, also, of other soluble salts, such as the various forms of "alkali" commonly found in the soils of the region, the toxicity of copper compounds is enormously lessened.

The investigations recorded in this publication include: (1) Observations upon the distribution of copper in mining wastes, in irrigating waters, in soils and soil waters, in the plants, and in the animal life of the region. (2) The development of accurate methods for the determination of minute amounts of copper in all situations where they may occur. (3) Plant cultural work with waters and in soils in the presence of varying proportions of copper and under varying conditions. (4) A careful analytical study of the results of such cultures in order to determine the symptoms of poisoning and the distribution of copper throughout poisoned plants; and to identify, if possible, the particular parts of plants and tissues injured by copper. (5) A physiological study of plant reactions with copper. (6) Field studies for the purpose of relating the results of laboratory investigations to the question of economic injury done by copper salts to irrigated crops.

By reason of interruptions due to other duties, it has required a long time to mature this investigation to the point where it seems sufficiently complete for publication. This delay, however, has given perspective to the work and, especially, opportunity to verify earlier conclusions as applied to field conditions.

The writer is indebted for painstaking analytical work to Messrs. R. G. Mead, Edward E. Free, Dr. W. H. Ross and

<sup>1</sup> See Bibliography, pp. 487-488, references 1, 8, 14, 15, 16, 19, 34, 51.

C. N. Catlin, associated with the Arizona Agricultural Experiment Station from time to time; and to the helpful advice of Dr. Howard S. Reed, of the University of California Graduate School of Tropical Agriculture, in connection with the physiological part of the work herein described. The publication, also, has been criticized to its advantage by Dr. C. B. Lipman of the University of California.

## DISTRIBUTION OF COPPER COMPOUNDS THROUGH- OUT THE CLIFTON-MORENCI AND GILA RIVER MINING AND IRRIGATION DISTRICTS

### SOURCES OF COPPER

The original source of the copper found in this district, according to Lindgren,<sup>2</sup> is a Cretaceous or early Tertiary intrusion of acidie porphyries to which, in the Clifton-Morenci district, all ore deposits may be finally referred. The original porphyries contain as little as 0.02 per cent of copper ore in the form of chalcopyrite. Under the influence of superheated waters emanating from the porphyry, this chalcopyrite, together with other metallic compounds, was carried out from the molten intrusive mass into adjoining strata and there deposited, especially along fissures, in the form of concentrated masses or veins of chalcopyrite and other minerals. Through erosion these deposits were afterward subjected to atmospheric oxidation, followed by downward percolation and a period of secondary enrichment due to numerous reactions mainly between the oxidized compounds of copper and other minerals present.

In limestones and shales, these processes resulted in the formation of oxidized ores containing azurite, malachite, chrysocolla, and cuprite. In porphyry, the main final result was chalcocite or copperglance, the principal constituent of the sulphide ores of the Clifton-Morenci district.

In general, therefore, the metasomatic changes associated, first, with superheated waters arising from the original intrusion of molten porphyry and, second, with meteoric waters percolating

<sup>2</sup> U. S. Geological Survey, Professional Paper No. 43, 1905.

downward with oxidizing effects through copper-bearing rocks, have brought copper from a concentration of possibly less than 0.02 per cent in the original porphyry through every degree of richness to the condition in some cases of pure copper.

#### PROCESSES BY WHICH COPPER IS ADDED TO THE WATER-SUPPLY

To a slight extent, drainage waters from the ore deposits and from the mines, containing considerable amounts of copper in solution, find their way downstream. But by far the larger part of the copper which gets into the irrigating supply is derived from the ores and tailings which, in the concentrators, on the dumps, and finally in the river itself, are subjected to the action of atmospheric oxygen, and water containing carbon dioxide and various salts in solution. The residual chalcocite in tailings from sulphide ores thus reacts with oxygen from the air and yields copper sulphate in solution. This, in turn, reacts with the excess of bicarbonate of lime ordinarily contained in the waters of the San Francisco and Gila rivers. The resulting basic carbonate of copper is notably soluble in water containing carbon dioxide and certain of the various salts commonly found in river waters. The residues of carbonates of copper in oxidized ores are directly dissolved in waters containing carbon dioxide and certain soluble salts.

Along with minute quantities of copper thus dissolved and carried forward, pass the solid residues discharged from the concentrators—solid wastes which find their way, unchanged, downstream and finally upon the soils of irrigated fields. At this point begins another and very important series of reactions between dissolved copper compounds and the soil, tending in general to withdraw copper from its solutions and precipitate it in the form of less harmful solid compounds. These are briefly referred to above and will be discussed more in detail further on in this paper. Opposing these precipitations of copper are those solvents which tend to maintain this metal in soluble form in small quantities in the soil. Chief of these is carbon dioxide, which is always present in agricultural soils in significant quantities. Of interest in this connection is the fol-



TABLE I  
SOLUBILITIES OF COPPER COMPOUNDS

Compound	Solvent	Cu dissolved, parts per million	Reference
Malachite $\text{CuCO}_3 \cdot \text{Cu}(\text{OH})_2$	Water containing 0.12% carbon dioxide	29.0-31.0	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1367
Precipitated basic copper carbonate	Pure water	1.5	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1370
Precipitated basic copper carbonate	Water containing 0.12% carbon dioxide	34.8	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1370
Precipitated basic copper carbonate	Water containing 0.13% carbon dioxide and 0.01% sodium chloride	36.0	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1371
Precipitated basic copper carbonate	Water containing 0.13% carbon dioxide and 1.0% sodium chloride	58.0	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1371
Precipitated basic copper carbonate	Water containing 0.12% carbon dioxide and 0.01% sodium sulphate	37.0	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1372
Precipitated basic copper carbonate	Water containing 0.12% carbon dioxide and 1.0% sodium sulphate	58.0	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1372
Precipitated basic copper carbonate	Water containing 0.12% carbon dioxide and 0.01% sod. carbonate	10.0	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1372
Precipitated basic copper carbonate	Water containing 0.12% carbon dioxide and 1.0% sod. carbonate	0.7	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1372
Precipitated basic copper carbonate	Water containing 0.12% carbon dioxide and 0.2% calcium sulphate	36.0	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1372
Precipitated basic copper carbonate	Water containing 0.12% carbon dioxide and 0.11% calc. carbonate	1.4	E. E. Free, Journ. Am. Chem. Soc., XXX, 9, p. 1372
Copper sulphide; $\text{CuS}$	Oxygen-free water	0.09	W. H. Ross, MSS
Chalcopyrite $\text{CuFeS}_2$	Pure water	measurable amounts	U. S. Geol. Survey Monograph XLVII, p. 1107
Chalcopyrite $\text{CuFeS}_2$	Sodic sulphide	Amt. not stated	U. S. Geol. Survey Monograph XLVII, p. 1106
Malachite	"Insoluble in water, slightly soluble in water charged with carbon dioxide."		Moissan 5, p. 167
Chrysocolla $\text{Cu Si O}_3 \cdot n \text{H}_2\text{O}$	"Somewhat soluble in water with carbon dioxide"		Lindgren, U. S. a. S. Prof. paper 43, p. 188
Cupric sulphide $\text{CuS}$	Water	1 to 950,000	Comey, Dict. Solu- bilities, p. 139
Cuprite $\text{Cu}_2\text{O}$	"Insoluble in water"		Comey, Dict. Solu- bilities, p. 137
Cupric oxide $\text{CuO}$	"Insoluble in water"		Comey, Dict. Solu- bilities, p. 137

lowing table of solubilities of various compounds of copper in different solvents, made up from different sources of information. The exact determinations of solubility by E. E. Free and W. H. Ross were made to obtain data needed in this investigation.

This table indicates that the carbonates and the silicate (chrysocolla) of copper, which are the compounds in which the metal must largely occur in the soil, are notably soluble in aqueous solutions of carbon dioxide.<sup>3</sup> Large amounts of sodium chloride and sodium sulphate increase the solubility of precipitated basic copper carbonate. In pure water, copper compounds, so far as observed, are but slightly soluble. Fluctuations in the content of carbon dioxide and of soluble salts in soil waters, and variations in the character of the soluble salts, are shown to affect the copper content of such waters.

In brief, the final effect upon plant roots of copper in the soil is the complex resultant of many opposing influences tending, on the one hand, to remove copper from solution, and, on the other, to maintain it in toxic soluble form. *Observations on the soil* usually fail to give satisfactory evidence as to the toxic or non-toxic effects to be expected from small percentages of copper that may be present. *Direct chemical and physiological studies of plants* afford much more satisfactory information. This mode of attack has been employed considerably in this investigation.

In view of the general tendency in nature to hinder the movements of copper in soils and to convert it into its insoluble forms, and independently of any tendency of the plant itself to assimilate or to reject copper, we should expect to find relatively small amounts of this element in plant tissues.

The following analytical determinations of copper in ores and tailings were made in samples carefully collected by the writer throughout the district studied. In all cases, the copper was determined electrolytically, manipulations of great delicacy having been developed for the determination of the minute amounts of copper often encountered. A full statement of the methods of preparing samples for analysis, and of determining

<sup>3</sup> Sullivan has shown that powdered silicates react with copper sulphate to withdraw copper from solution; and that this copper will then be redissolved by a solution of potassium sulphate. U. S. Geol. Survey, Bull. 312, 1907.

copper in ores, tailings, waters, soils, and organic materials, is to be found under "Methods of Analysis" in the appendix to this paper. For convenience in comparing widely variable amounts in the samples examined, the copper content is given in parts per million of substance. Parts per million may be reduced to percentages by moving the decimal point four places to the left. For instance, 11,600 parts per million is equal to 1.16 per cent.

TABLE 11

## COPPER IN ORES AND TAILINGS FROM THE CLIFTON-MORENCI MINING DISTRICT

Sample No and date	Description of sample	Condition and weight taken, grams	Cu found, grams	Cu parts per million
3491	One day's run of sulphide ore from	1		
May 23, '04	from A. C. Co.'s mill in Clifton	air-dry	.03195	31,950
3303	Sulphide tailings, point of discharge	1		
May 23, '04	from A. C. Co.'s mill, Clifton	water-free	.00935	9,350
3438	Sulphide tailings, point of discharge	1		
June 28, '05	from A. C. Co.'s mill, Clifton	water-free	.0116	11,600
3499	Sulphide tailings at Clifton coming	1		
June 28, '05	from Longfellow mill	water free	.00725	7,250
D. C. Co.'s Records	Fine sulphide tailings at Morenci			10,000
May 20, '04				
3492	One day's run of oxidized ore from	1		
May 23, '04	A. C. Co.'s mill at Clifton	air-dry	.0553	55,300
3304	Oxidized tailings, point of discharge	1		
May 23, '04	from A. C. Co.'s mill, Clifton	water-free	.0225	25,500
3439	Oxidized tailings, point of discharge	1		
June 28, '05	from A. C. Co.'s mill, Clifton	water-free	.0268	26,800
3500	Oxidized tailings, point of discharge	1		
June 27, '05	from Shannon C. Co.'s mill, Clifton	water-free	.0114	11,400
3309	Milky sediments, pure tailings from	2		
May 26, '04	Montezuma Canal, Solomonville	water free	.01725	8,625
3486	River sediments with tailings from	2		
June 11, '05	Montezuma Canal, Solomonville	water-free	.0067	3,350
3737	High river sediments with muddy			
Feb. 22, '07	tailings from Montezuma Canal, Solomonville	15.8 water-free	trace	trace
6342	Floodwater sediments from Monte-			
Mar. 4, '16	zuma Canal, Solomonville; tail- ings from Mogollon	.5899	.000085	53

A point of interest to both mine owners and farmers in table II is the large proportion of copper that was discarded with tailings at the time the samples were taken. This loss, so far as these figures show, may amount to almost one-third of the copper

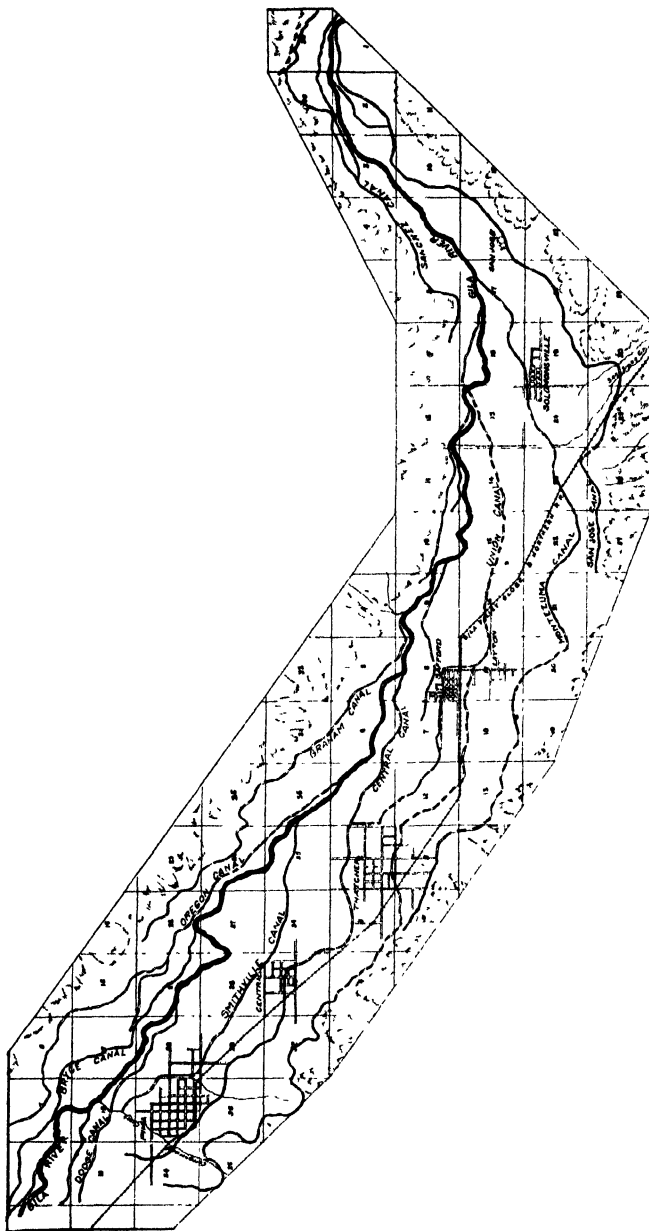


Fig. 2.—Detail map of the Gila River irrigation district, in Graham County, Arizona.

in low-grade sulphide ores (No. 3491, No. 3303), and to nearly one-half in the richer oxidized ores (No. 3492, No. 3304, No. 3439). By far the larger portion of tailings produced, however, are from low-grade sulphide ores, the wastes from which therefore predominate, formerly imparting to river waters the whitish

TABLE III

DISSOLVED COPPER IN RIVER, IRRIGATING, AND GROUND WATERS BELOW THE  
CLIFTON-MORENCI DISTRICT

Sample No. and date	Description of sample	Condition and amount taken in cc.	Cu found, grams	Cu p.p.m.
3438	Water mixed with sulphide tailings			
June 28, '05	from A. C. Co.'s mill, Clifton	500	.0009	1.80
3439	Water mixed with oxidized tailings			
June 28, '05	from A. C. Co.'s mill, Clifton	500	.0018	3.60
3309	Montezuma Canal water at Solo-			
May 26, '04	monville; slight rise in river	2000	.0016	.80
3486	Montezuma Canal at Solomonville,			
June 11, '05	small flood	6000	.0015	.25
3622	Montezuma Canal at Solomonville,			
June 25, '06	head waters clear	9000	.00095	.11
3737	Montezuma Canal at Solomonville,			
Feb. 22, '07	medium flood	14000	.0403	2.88
Tailings shut out of water supply May 1, 1908.				
4011	Water from Montezuma Canal at			
Jan. 3, '09	Solomonville	4000	.00031	.08
4029	Montezuma Canal at Solomonville	3700	.0003	.08
Apr. 12, '09				
6342	Montezuma Canal at Solomonville,			
Mar. 4, '16	high water	1000	.00003	.03
3986	Water from C. & A. Ditch, Bisbee			
Jan. 2, '09	mine waters	3500	.00039	.11
Records of				
Cananea				
C. C. Co.	Water from creek below concen-			
Jan. 4, '14	trator			2.1
3504	Water from Geo. Olney's well, 30 ft.			
Aug. 19, '05	deep, east of Safford, under			
	Montezuma Canal	7000	.0037	.53
4012	Water from Wilson's well, one-half		less	less
Jan. 3, '09	mile west of Solomonville under		than	than
	San Jose Canal	3500	.00001	.003
3526	Water from University well, Tuc-			
	son, 95 ft. deep, tapping Rillito			
	underflow	7000	none	none

appearance characteristic of this material. It is of interest to note in this connection that in one instance observed the tailings almost completely maintained their richness in copper between

Clifton and Solomonville. At Clifton, May 23, 1904, the principal discharge of sulphide tailings (3303) was observed carrying 0.93 per cent of copper. At Solomonville, three days later, the Montezuma ditch-water sediments (No. 3309), mostly of this same material, carried 0.86 per cent of copper, indicating the persistence with which the copper accompanies the wastes, with which it is associated, downstream and upon underlying irrigated lands.

TABLE IV  
COPPER IN SOILS IRRIGATED WITH TAILINGS WATERS

Sample No. and date	Description of sample	Condition and weight taken, grams	Cu found, grams	Cu p.p.m.
3435 May 25, '04	Top 5 in. sedimentary soil (Fred Thorstison), upper end alfalfa field west of Safford, under Montezuma Canal	96.7 water-free	.020	207
3434 June 10, '05	Top 5 in. sedimentary soil (Geo. Olney), upper end alfalfa field east of Safford, under Montezuma Canal	96.8 water-free	.0199	205
3501 Aug. 19, '05	Soil in place at 4 ft. depth beneath No. 3434	96.2 water-free	.0021	22
3436 June 25, '05	Top sedimentary soil (Wm. Gillespie), upper end of test alfalfa field west of Solomonville, under Montezuma Canal	96.7 water-free	.0192	199
3437 June 25, '05	Soil in place, no sediments at surface of lower end of field near No. 3436	94 water-free	.0028	30
3502 Aug. 19, '05	Soil in place at 4 ft. depth beneath No. 3437	96.1 water-free	.001	10
2381 June 5, '00	Surface 12 in. from garden near Pima, Ariz., beyond tailings deposits	100 air-dry	faint trace	trace
3522 Oct. 25, '05	Top 4 in. sedimentary soil upper end of alfalfa field, Station farm near Phoenix, under Grand and Maricopa canals	95 water-free	.0003	3
3521 Oct. 25, '05	Deep soil, no sediments, Station farm near No. 3522	95 water-free	.0003	3
2763 Nov. 11, '01	Surface 12 in. from orange orchard north of Phoenix, under Arizona Canal	100 air-dry	faint trace	trace
1890 Apr. 20, '01	Surface 15 in. from cultivated field west of Tempe, under Tempe Canal	100 air-dry	faint trace	trace
2830 Jan. 19, '00	Surface 12 in. from orange orchard northeast of Phoenix, under Arizona Canal	100 air-dry	none	none

Table III is of interest because it reveals quantities of dissolved copper in irrigating and in ground waters sufficient, under proper conditions, in water cultures, to produce toxic effects upon plants.<sup>4</sup> It is noteworthy, however, that, following the order of the court, effective May 1, 1908, prohibiting the introduction of tailings into the water-supply, the amount of dissolved copper in Montezuma canal waters greatly decreased, due to the decrease in quantity of sulphides whose oxidation affords the supply of dissolved copper. Other water-supplies also are found to contain similar amounts of copper, as the Calumet and Arizona mine waters, used for irrigation below Bisbee. As stated above, however, in the soil itself the toxic action of such copper solutions is enormously decreased. Naturally, the question arises as to the possibility of toxic effects in using such waters upon cultivated soils. This is discussed on subsequent pages. The proportions of copper (0.003 to 0.53 parts in 1,000,000 of water) found in the drainage beneath this irrigated district indicate that not all of the copper applied in irrigation remains in the soil. University well water at Tucson was observed to be free from this element.

Soils Nos. 3435, 3434, and 3436 show maximum amounts of copper, inasmuch as they are composed to a considerable extent of tailings. The soils in place beneath these sediments, Nos. 3501 and 3502, contain much less, yet noticeable amounts of copper, most of which is retained where it first comes in contact with the top soil. It is of interest to note that the surface sediments and the deep soils of the Experiment Station farm near Phoenix, Arizona, irrigated from an entirely different watershed, also contain small but weighable amounts of copper. This was probably derived from mines at Globe and Jerome, Arizona, whose wastes have found their way into the drainage which supplies irrigation for Salt River Valley. The quantities observed, however, three parts copper per million of soil, are negligible. Other soils from Salt River Valley also show traces of copper.

<sup>4</sup> See Bibliography, p. 487, references 5, 18.

TABLE V

## MISCELLANEOUS SOILS UNAFFECTED BY MINING DETRITUS

Sample No. and date	Description of sample	Condition and weight taken, grams	Cu found, grams	Cu p.p.m.
2375 June 5, '00	Surface 12 in. from new ground near Safford, recently placed under Montezuma Ditch	100 air-dry	none	none
2253 Jan. 3, '00	Surface 12 in. university ground, Tucson	100 air-dry	none	none
3503 May 9, '05	Surface 12 in. virgin unirrigated soil, Colorado Valley bottom near Yuma	100 air-dry	none	none

These determinations, made in widely separated localities, indicate the absence of copper in soils which are not immediately under the influence of mining detritus.

TABLE VI

## COPPER IN VEGETATION FROM UPPER GUA VALLEY FARMS

Sample No. and date	Description of sample	Condition and weight taken, grams	Cu found, grams	Cu p.p.m.
3505 Aug. 19, '05	Alfalfa, before blooming, from upper end of Geo. Olney's field east of Safford, under Monte- zuma Ditch	1206 air-dry	.0062	5.10
3512 Aug. 19, '05	Alfalfa from bale grown in Lay- ton (M. B. Steele) under Monte- zuma ditch	1359 air-dry	.0077	5.70
3507 Aug. 20, '05	Corn in bloom, leaves only, grown in Layton (Jas. Welker), under Montezuma Ditch	545 air-dry	.0033	6.10
3509 Aug. 19, '05	Wheat from stack, stalk and grain, grown in Layton (M. B. Steele), under Montezuma Ditch	1125 air-dry	.0027	2.40
3513 Sept. 19, '05	Mistletoe, growing on willow 25 ft. above ground, one mile east of Safford, under Montezuma Ditch	1245 air-dry	.0094	7.60
3739	Alfalfa seed, crop of 1906, grown near Pima under Smithville Ditch	782 water-free	.0026	3.33
3741	Alfalfa seed (Wm. Gillespie), crop of 1906, grown near Solomonville, under Montezuma Ditch	843 water-free	.0023	2.72
3780	Shelled corn, crop of 1906, grown at Solomonville, under Monte- zuma Ditch	932 water-free	.0004	.43
3740	Shelled corn, crop of 1906, grown at Solomonville, under Monte- zuma Ditch	874 water-free	.0008	.73
3738	Shelled corn, crop of 1906, grown near Pima, under Smithville Ditch	1092 water-free	trace	trace



The prevalence of small amounts of copper in vegetation throughout this locality is shown by the figures in table VI. Samples of corn and alfalfa contained comparable quantities of copper, which, however, were exceeded by the amount found in a sample of mistletoe growing on a willow fully twenty-five feet above the ground. This is due chiefly to the perennial character of mistletoe which, therefore, has more time to accumulate copper. It is interesting to note also that seeds of alfalfa and corn contain less copper than corresponding foliage. Corn leaves were observed to contain 6.1 parts of copper per million parts of air-dry substance, while grain from the same locality contained from 0.73 to 0.43 parts. Alfalfa seed contained about one-half as much copper as the stalks and leaves, while wheat hay carrying a large proportion of grain showed a low proportion of copper. These facts are probably connected with transpiration.

TABLE VII

## COPPER IN VEGETATION FROM OTHER LOCALITIES

Sample No. and date	Description of sample	Condition and weight taken, grams	Cu found, grams	Cu p.p.m.
		2109		
3508	Alfalfa hay, station farm near	air-dry	.0021	1.00
Aug. 25, '05	Phoenix (two samples)	1408		
		air-dry	.0031	2.20
3516	Alfalfa, before blooming, station	1106		
Oct. 25, '05	farm near Phoenix	air-dry	.0011	1.00
3515	Alfalfa hay, Colorado bottom,	1262		
Oct. 4, '05	Yuma date orchard	air-dry	none	none
3517	Barley hay, station farm near	1304		
May, 1905	Phoenix	air-dry	.0002	.15
3518	Corn, leaves only, station farm near	595		
Oct. 25, '05	Phoenix	air-dry	.0005	.84
3519	Corn, leaves only, grown on Rillito	284		
Oct. 14, '05	near old Fort Lowell	air-dry	.0018	6.30
3529	Corn, leaves and bloom, same field	1132		
Dec. 27, '05	as No. 3519	air-dry	.0015	1.32
3520	Mistletoe from cottonwood 30 ft.			
Oct. 14, '05	above ground, old Fort Lowell, near Tucson	1160		
		air-dry	.001	.85
3989	Young (5 mos. old) alfalfa roots			
Dec. 31, '08	from C. & A. ranch irrigated with mine waters containing cop- per, from Bisbee	2.12		
		air-dry	.0001	47.00
3990	Corn roots from C. & A. ranch irri- gated with mine waters contain- ing copper, from Bisbee	16.7		
		air-dry	.00025	15.00

which is maximum in leaves and quantitatively small in the fruiting parts of a plant. Additional evidence of this fact is shown in poisoned corn plants, which are discussed on a subsequent page.

Comparing the data of table VII with those of table VI, it is evident that, excluding corn and alfalfa irrigated with C. & A. mine waters, in every case except that of one sample of corn from old Fort Lowell (No. 3519) the copper in crops grown on Gila Valley farms is much in excess of that in plants coming from elsewhere for the same classes of material. The presence of appreciable amounts of copper in samples of alfalfa, corn, barley, and mistletoe also accords with the fact that the soils in which they were grown receive the drainage from copper-bearing watersheds. The one exception, at Yuma (No. 3515) where no trace of copper could be found either in alfalfa or in soil (No. 3503), indicates that these alluvial river deposits, which have been subjected annually to the leaching action of enormous quantities of flood waters, have been prevented from accumulating appreciable quantities of copper.

#### COPPER IN THE FLESH AND BONES OF A PIG

In order to follow the copper as far as possible in its trans-migrations, a five-months-old pig that had been born and brought up in an alfalfa pasture near Solomonville under the Montezuma Ditch, was killed and portions of the flesh and bones were taken for examination, with the following results:

Sample No. and date	Description of sample	Condition and weight taken, grams	Cu found, grams	Cu p.p.m.
3779		917		
May 7, '07	Liver, heart, and rib meat	fresh	.0053	5.78
3778		998		
May 7, '07	Ribs and rib meat	fresh	.00006	.06

The largest amount of copper was found in portions of liver, heart and rib meat, only minute amounts being present in the bony material. In this connection, it is stated that about two parts of copper have been observed in one million parts of human liver; ten parts in human kidneys, and as much as fifty parts

in sheep's liver.<sup>5</sup> Human food, however, is commonly contaminated with copper compounds, which account for its presence in the human body.

*In brief*, the observations detailed above have shown the successive positions of copper in the original ores of the Clifton-Morenci district; in the tailings wastes from these ores, in suspension and in solution in river waters exposed to milling operations; in soils irrigated with these waters; in the ground waters beneath these soils; in vegetation growing upon them; and even in the animal life of the region. It is of interest to observe, first, the concentration through natural processes of small amounts of copper in the original rocks into the form of rich ores; and, second, the reversal, through human agencies, of this process, and the dilution of copper values till, in vegetation and in animal life, but traces of the metal can be detected.

## DISTRIBUTION OF COPPER IN PLANTS WITH ROOT SYSTEMS EXPOSED TO COPPER COMPOUNDS

### CORN PLANTS GROWN IN SOILS CONTAINING COPPER

In order to determine accurately the distribution of copper throughout a typical crop plant, thereby locating if possible the points at which injury may occur from copper compounds in the soil, three lots of corn plants were examined in detail. Two of these were grown (August 3 to November 13, 1907) in pots containing thirty-eight pounds of sandy loam soil very thoroughly mixed with 0.01 and 0.025 per cent of copper in the form of freshly precipitated copper carbonate ( $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ ), made by mixing equivalent amounts of copper sulphate and sodium carbonate. The third was grown in soil containing 0.05 per cent of copper in the form of finely pulverized chalcocite.

The samples were harvested with care to prevent contamination with copper dust; the root portions being washed in copper-free water saturated with carbon dioxide until the washings contained no trace of copper. Determinations of copper, as

<sup>5</sup> Blyth, *Poisons*, fourth edition, pp. 640-641.

usual, were made as shown under "Methods of Analysis" (see Appendix herewith). Following are the tabulated results:

TABLE VIII  
ELEVEN STALKS OF CORN GROWN IN SOIL CONTAINING 0.01 PER CENT  
COPPER AS  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$  (1907)

No.	Plant part	Weight of sample, grams	Cu found, grams	Cu p. p. m.
3869p	Lower six nodes, 24 in. long	43.4	.00012	3.00
3869q	Basal sheaths of leaves from lower six nodes	23.2	.0001	4.00
3869r	Blades of leaves from lower six nodes	33.1	.00029	9.00
3869s	Upper four-seven nodes, 24 in. long	24.2	.00017	7.00
3869t	Basal sheaths of leaves from upper nodes	20.6	.00013	6.00
3869u	Blades of leaves from upper nodes	19.6	.00024	12.00
3869v	Rudimentary ears	11.8	.0001	9.00
3869	Whole top portions	175.9	.00115	6.50
3868	Roots	10.6	.00161	152.00

TABLE IX  
TEN STALKS OF CORN GROWN IN SOIL CONTAINING 0.025 PER CENT  
COPPER AS  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$  (1907)

No.	Plant part	Weight of sample, grams	Cu found, grams	Cu p. p. m.
3865a	Five lower nodes, 14.4 in. long	17	.00024	14.00
3865b	Basal 12 in. of leaves and sheaths from five lower nodes	17.2	.00037	22.00
3865c	Terminal 14 in. of leaves from five lower nodes	14.4	.00047	33.00
3865d	Upper five-seven nodes, 14 in. long, including tassels and ears	9.2	.00017	19.00
3865e	Leaves from same	19.2	.00035	18.00
3865	Whole top portions	77	.0016	21
3866	Roots	9.2	.0067	728

TABLE X  
FOUR STALKS OF CORN GROWN IN SOIL CONTAINING 0.05 PER CENT  
COPPER AS  $\text{Cu}_2\text{S}$ . (1908)

No.	Plant part	Weight of sample, grams	Cu found, grams	Cu p.p.m.
3968p	Lower six nodes	11.5	.00010	9.00
3968q	Basal sheaths from lower six nodes	7.5	.00008	11.00
3968r	Blades from do.	15.7	.00021	13.00
3968s	Upper five-six nodes	3.5	.00004	11.00
3968t	Basal sheaths from upper five-six nodes	5.2	.00007	13.00
3968u	Blades from do.	4.9	.00010	20.00
3968v	Rudimentary ears	3.4	.00005	15.00
	Whole top portions	51.7	.00065	12.50
3978a	Fine roots	3.23	.00081	251.00
3978b	Coarse roots	2.91	.00024	83.00
	whole root system	6.14	.00105	171.00

In all of the corn samples shown above, the copper content of root systems is very much greater than that in the top portions of the plants, amounting to twenty-three times, thirty-four times, and thirteen times as much, respectively. In the aerial parts of all samples copper increases slightly but uniformly towards the upper and outer portions of the plants. This must be an effect of transpiration, by which copper in solution is carried to the terminal portions of the plant and there deposited. The fine roots of one sample were found to contain about three times as much copper as the coarse roots—a fact which can be explained by the greater proportion of absorbing surface to weight in small roots.

With reference to toxic effects, the culture in 0.01 per cent copper carbonate showed only a faint yellow striping of leaves, with no checking of growth. The 0.025 per cent culture gave leaves which were strongly striped with yellow, and the total growth reduced to less than one-half. Toxic effects evident in the top portions of this culture are manifestly to be associated mainly with the greatly increased copper content of its roots, since total amounts of copper in the top portions remain small. The 0.05 per cent culture of copper in the form of  $\text{Cu}_2\text{S}$ , or finely powdered chalcocite, showed only faint toxic effects in the tops. The following summary indicates the relation between toxic effects and copper content of materials.

Culture	Condition	Cu in tops p.p.m.	Cu in roots p.p.m.	Ratio
Copper carbonate, 0.01% Cu (precipitated)	Leaves faintly striped normal weight	6.50	152.00	1:23
Copper carbonate, 0.025% Cu (precipitated)	Leaves strongly striped three-fourths yellow, half weight	21.00	728.00	1:34
Copper sulphide, 0.05% Cu	Faintly striped leaves, normal weight	12.50	171.00	1:13

In this table a general relation is shown between the toxic effects in the aerial portions of the plant, and the amounts of copper in root systems; but as to the soils employed toxic effects are influenced both by amounts and character of copper compounds present, as is shown further on following pages.

In view of the fact that the small increase of copper in the carbonate cultures, from 0.01 to 0.025 per cent, caused severe toxic effects attended by an increase of copper in root systems from 152 to 728 p.p.m. of dry matter, it seemed desirable to investigate thoroughly the quantitative relations between the copper in roots and the toxic effects as shown in vegetative growth. It was expected in this way to find a means of determining whether a plant contained an injurious or killing dose of copper, just as, analogously, killing doses of poisons in animals may be ascertained. With this end in view cultures of corn, beans, and squashes were grown in water, in pots of soil and in garden plots; and roots and top portions were examined quantitatively for copper.

In preparing samples of roots for analysis, washing with 4 per cent hydrochloric acid was carried out with water cultures, but most of the samples were prepared by washing with large quantities of copper-free water saturated with carbon dioxide, until the washings showed no trace of copper. By still a third method the soil adhering to a sample was analyzed for copper, the ash was then determined and assumed to be soil, and a corresponding amount of copper subtracted from the total found. For details see "Methods of Analysis." All of these methods undoubtedly give conservative figures for copper in root systems inasmuch as solvents not only remove externally adhering compounds but may also gradually act upon the copper content of

root systems. The acid-wash and soil-correction methods give severely minimum results. The carbon dioxide wash used in the majority of analyses is laborious but more satisfactory.

### WATER CULTURES (1907)

Cultures of corn, beans, and squash were grown in University of Arizona well-water containing 250 p.p.m. of soluble solids. From 0.03 to 3.0 parts of copper as precipitated carbonate dissolved in carbon dioxide were used in making cultures and the resulting growths of tops and roots were divided into the worst-poisoned and least-poisoned portions, for determinations of copper.



Fig. 3.—Corn cultures, series 121-62, grown in University of Arizona well water, containing from .03 to 3. parts per million of copper as basic carbonate ( $\text{Cu}(\text{OH})_2\text{CuCO}_3$ ).

*Series Corn 121-62.*—Grown in well water containing Cu as  $\text{Cu}(\text{OH})_2\text{CuCO}_3$  as follows: check, 3.0, 1.0, 0.8, 0.5, 0.3, 0.1, 0.08, 0.05, and 0.03 p.p.m. Cu. December 1–February 27, 1907. Series divided into two portions:

a. Plants not badly poisoned; roots growing; tops showing Cu effects; 0.1, 0.08, 0.05, 0.03 cultures. (Nos. 3694, 3693.)

b. Plants badly poisoned; root growth arrested; tops living; 3.0, 1.0, 0.8, 0.5, and 0.3 cultures. (Nos. 3692, 3691.)

*Series Beans 121-66.*—Grown in well water containing Cu as  $\text{Cu}(\text{OH})_2\text{CuCO}_3$  as follows: check, 3.0, 1.0, 0.8, 0.5, 0.3, 0.1, 0.08, 0.05, 0.03 p.p.m. Cu. December 9–February 27, 1907. Series divided into two portions:

a. Least poisoned plants; roots nearly normal, tops normal; 0.3, 0.1, 0.08, 0.05, 0.03 cultures. (Nos. 3702, 3697.)

*b.* Worst poisoned plants; roots badly affected, tops less affected; 3.0, 1.0, 0.8, and 0.5 cultures. (Nos. 3700, 3699.)

*Series Squash 121-66.*—Grown in well water containing Cu as  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$  as follows: check, 3.0, 1.0, 0.8, 0.5, 0.3, 0.1, 0.08, 0.05, and 0.03 p.p.m. Cu. December 10–February 27, 1907. Series divided into two portions:

*a.* Least poisoned plants; roots growing; tops strong; 0.3, 0.1, 0.08, 0.05, and 0.03 cultures. (Nos. 3698, 3701.)

*b.* Worst poisoned plants; roots dead or nearly so; tops badly affected; 3.0, 1.0, 0.8, and 0.5 cultures. (Nos. 3696, 3695.)

TABLE XI  
COPPER CONTENT OF PLANTS IN WATER CULTURES

No.	Series	Condition of sample	Dry matter in grams	Cu found, grams	p p m	
					Cu dry matter	
					tops	roots
3694	Corn, .1, .08, .05, .03	Tops affected	6.3	.00009	14.30	
3693	Corn, .1, .08, .05, .03	Roots growing	2.3	.000236		102.60
3692	Corn, 3., 1., .8, .5, .3	Tops living	4.8	.000056	11.70	
3691	Corn, 3., 1., .8, .5, .3	Roots arrested	2.8	.000572		204.30
3702	Beans, .3, .1, .08, .05, .03	Tops normal	9.4	.000198	21.10	
3701	Beans, .3, .1, .08, .05, .03	Roots growing	2.6	.000157		60.40
3700	Beans, 3., 1., .8, .5	Tops affected	6.6	.000204	30.90	
3699	Beans, 3., 1., .8, .5	Roots badly affected	1.6	.000494		308.80
3698	Squash, .3, .1, .08, .05, .03	Tops strong	10.4	.000333	32.00	
3697	Squash, .3, .1, .08, .05, .03	Roots nearly normal	.6	.000087		145.00
3696	Squash, 3., 1., .8, .5	Tops badly affected	3.6	.000092	26.00	
3695	Squash, 3., 1., .8, .5	Roots dead	.2	.000058		290.00

It is noteworthy, in this series, that the amounts of copper found in roots that still retain the power of growth average about 103 parts in one million of dry matter, as compared with 268 parts in dead roots whose protoplasm is presumably killed as an effect of copper. Badly poisoned roots in every instance show a great excess of copper over those less affected. The tops, on the other hand, do not show copper in proportion to the amounts in the roots, averaging the same amount of copper in badly poisoned (22.9 p.p.m.) and in slightly poisoned (22.5 p.p.m.) plants. Corn was observed to be distinctly more sensitive to copper in water culture than either squash or beans, as was



shown by the method of measuring growth of root tips marked with India ink, and noting points at which growth was retarded (*R*) and arrested (*A*).

TABLE XII

SHOWING POINTS AT WHICH ROOTS WERE RETARDED OR ARRESTED IN GROWTH

Cultures in Cu, in well water, parts per million .....		.03	.05	.08	.1	.3	.5	.8	1.	3.
Corn .....				R					A	
Beans .....					R				A	
Squash .....					R				A	

Photographs of the three series also indicate an earlier retardation of corn root development than of bean or squash root development; and show additionally that the top portions of cultures are not damaged in proportion to the root systems.

## TOXICITY OF COPPER SOLUTIONS TO PLANT ROOTS IN WATER CULTURE

In order to gain some indication of effects in water culture of copper salts upon plants, several series of plants were grown under varying conditions, and effects observed of the kind of copper salts employed, strength of solution used, the kind of plant, and the effects of other salts present.

Solutions were made in water free from copper, twice distilled; or, where permissible, University of Arizona well water, copper-free. The series were arranged, usually, to carry 0.01, 0.03, 0.05, 0.08, 0.1, 0.3, 0.5, 0.8, 1.0, 3.0, and 5.0 parts copper per million of water. The cultures were made in 600-c.c. bottles, covered with pasteboard squares saturated with hot paraffin and perforated with three holes for plant seedlings held in place by cotton.

Effects upon cultures were judged by elongation of roots determined by the usual method of marking with India ink 5 mm. back of root tips and noting growth after twenty-four hours. Corn, beans, and squash were the plants employed and the points

particularly noted were those at which growth was retarded and at which it was arrested.

Table XIII gives the data condensed from the experimental records:

TABLE XIII (a)  
TOXIC EFFECTS OF COPPER UPON ROOTS OF WATER CULTURES  
First experiment (1905)

Culture	Copper salt employed	Kind of water	Cu in solution	
			Growth retarded between, p.p.m.	Growth arrested between, p.p.m.
Beans	$\text{CuSO}_4$	Distilled		.25—1.25
Beans	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$	Well		.57—5.7
Cantaloupes	$\text{CuSO}_4$	Distilled		less than .25
Cantaloupes	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$	Well		.57—5.7

Indicating lessened toxicity in well water.



Fig. 4.—Bean cultures (eighth exp.), showing effects of varying concentrations of copper in distilled water and in solutions of mixed salts. S, salt solutions; D, distilled water; W, no copper, and .05 to 3. parts per million of copper.

TABLE XIII (b)  
Eighth experiment (1905)

Corn	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$	Salt solution*	.3 — .5	.8 — 1.
Corn	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$	Distilled	less than .01	.1 — .3
Beans	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$	Salt solution*	.1 — .5	.8 — 1.
Beans	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$	Distilled	.1 — .3	.5 — .8
Squash	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$	Salt solution*	.1 — .5	.8 — 1.
Squash	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$	Distilled	.1 — .3	.3 — .5

Showing lessened toxicity in salt solution.

\* NaCl 64 pts.

$\text{Na}_2\text{SO}_4$  2

$\text{CaSO}_4$  7.3

Univ. well

water salts 26.1

Total 100 pts. per 100,000.

TABLE XIII (c)  
Fifth experiment (1905)

Corn	CuSO <sub>4</sub>	Distilled	.01— .05	.1 — .5
Corn	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	Distilled	.01— .05	.1 — .5

Indicating equal toxicity of Cu as sulphate and as carbonate, and (compare second, fourth and eighth experiments) great toxicity to corn in distilled water.

TABLE XIII (d)  
Third experiment (1905).

Beans	CuSO <sub>4</sub>	Distilled		.1 — .3
Beans	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	Distilled		.1 — .3

Toxicity to beans of Cu as sulphate and as carbonate was the same.

TABLE XIII (e)  
Seventh experiment (1905)

Squash	CuSO <sub>4</sub>	Distilled	.01— .05	.1 — .5
Squash	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	Distilled	.01— .05	.1 — .5

Toxicity to squash of Cu as sulphate and as carbonate was the same.

TABLE XIII (f)  
Second experiment (1905)

Culture	Copper salt employed	Kind of water	Cu in solution	
			Growth retarded between, p. p. m.	Growth arrested between, p. p. m.
Corn	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	Well	.1 — .3	.8 — 1.
Beans	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	Well	.1 — .3	.8 — 1.

Toxicity of copper as Cu(OH)<sub>2</sub>.CuCO<sub>3</sub> to corn and beans was the same.

TABLE XIII (g)  
Fourth experiment (1905)

Corn	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	Well	.05— .08	.8 — 1.
Squash	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	Well	.1 — .3	.8 — 1.

Corn was somewhat more sensitive to copper as Cu(OH)<sub>2</sub>.CuCO<sub>3</sub> than squash.

TABLE XIII (h)  
Sixth experiment (1905)

Beans	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	Well	.1 — .3	1. — 3.
Squash	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	Well	.1 — .3	.8 — 1.

Beans and squash were about equally sensitive to copper as Cu(OH)<sub>2</sub>.CuCO<sub>3</sub>.

These experiments, which are not stated in complete detail here, indicate quite clearly:

1. That the toxic effects of copper are less in the presence of the salts ordinarily contained in well waters than in distilled-

water solution. This fact indicates that the toxicity of copper salts in the presence of soil-water solutions is probably minimized.

In all cases it was observed that root growth was much more vigorous in salty than in distilled water, where no copper was used. Lessened toxicity of copper in salty solutions may therefore *in part* be due to greater vigor and resistant qualities of plant cells grown in such solutions.

2. Copper appears to be equally toxic as sulphate or as basic carbonate.

3. Corn is probably more sensitive to copper salts than is squash or beans.

#### STIMULATION EFFECTS IN WATER CULTURES

In view of the debated question as to stimulation of plant growth by minute amounts of copper salts, it is of interest to observe that, quite consistently, the most vigorous root growth is associated with concentrations of from 0.01 to 0.1 parts per million of copper, as shown by details from cultures described on previous pages.

TABLE XIV (a)

STIMULATION EFFECTS OF COPPER UPON ROOTS OF PLANTS IN WATER CULTURES

*Corn roots* grown in well water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$

Cu p.p.m.	Elongation 48 hrs	Condition
check	23.4 mm.	Tops of plants showing
.01	27.3	increased growth at .08
.03	17.6	and .1 p.p.m.
.05	17.3	
.08	19.4	
.1	18.5	

Showing stimulation at .01 p.p.m.

TABLE XIV (b)

*Bean roots* grown in well water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$

Cu p.p.m.	Elongation 48 hrs.	Condition
check	2.5 mm.	Tops of plants in .08
.01	2.2	and .1 cultures higher
.03	4.7	than in .05, .03, .01, and
.05	2.8	check.
.08	2.5	
.1	2.9	

Showing stimulation at .03 p.p.m.

TABLE XIV (c)

*Corn roots grown in well water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$* 

Cu p.p.m.	Elongation 48 hrs.	Condition
check	14.2 mm.	vigorous
.03	17.3	most vigorous
.05	14.4	most vigorous
.08	9.6	retarded
.1	10.3	retarded

Showing stimulation at .03 p.p.m.

TABLE XIV (d)

*Squash roots grown in well water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$* 

Cu p.p.m.	Elongation 48 hrs.	Cu p.p.m.	Elongation 48 hrs.
check	12.0 mm.	.08	12.3
.03	10.7	.1	13.2
.05	10.0		

Showing no stimulation at these concentrations.

TABLE XIV (e)

*Bran roots grown in well water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$* 

Cu p.p.m.	Elongation 48 hrs.	Condition
check	2.4 mm.	Tops strong through-
.03	3.1	out, showing stimula-
.05	4.5	tion at .03, .05, and .1
.08	2.8	
.1	3.	

Showing stimulation at .05 p.p.m.

TABLE XIV (f)

*Squash roots grown in well water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$* 

Cu p.p.m.	Elongation 48 hrs.	Cu p.p.m.	Elongation 48 hrs.
check	13.1 mm.	.08	7.6 mm.
.03	7.4	.1	9.7
.05	8.8	.3	3.7

Not showing stimulation consistently.

TABLE XIV (g)

*Corn roots grown in well water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$* 

Cu p.p.m.	Elongation 48 hrs.	Cu p.p.m.	Elongation 48 hrs.
check	9.8 mm.	.1	13.5 mm.
.01	13.8	.3	10.
.05	17.5	.5	3.8

Showing strong stimulation .01 to .1 mm.

TABLE XIV (h)

*Corn roots* grown in distilled water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ 

Cu p.p.m.	Elongation 48 hrs.	Cu p.p.m.	Elongation 48 hrs.
check	27.6 mm.	.05	5.7 mm.
.01	1.8	.1	2.7

No stimulation; eccentric results.

TABLE XIV (i)

*Bean roots* grown in well water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ 

Cu p.p.m.	Elongation 48 hrs.	Cu p.p.m.	Elongation 48 hrs.
check	2. mm.	.1	6. mm.
.05	6.	.5	.8

Showing stimulation at .05 to .1 p.p.m.

TABLE XIV (j)

*Bean roots* grown in distilled water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ 

Cu p.p.m.	Elongation 48 hrs.	Cu p.p.m.	Elongation 48 hrs.
check	3. mm.	.1	3.1 mm.
.01	3.	.3	1.6
.05	3.7		

Showing no stimulation.

TABLE XIV (k)

*Squash roots* grown in well water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ 

Cu p.p.m.	Elongation 48 hrs.	Cu p.p.m.	Elongation 48 hrs.
check	2.4 mm.	.1	5.7 mm.
.05	4.9	.5	.4

Showing stimulation at .05 to .1 p.p.m.

TABLE XIV (l)

*Squash roots* grown in distilled water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ 

Cu p.p.m.	Elongation 48 hrs.	Cu p.p.m.	Elongation 48 hrs.
check	3.3 mm.	.05	3.1 mm.
.01	3.3	.1	2.1

Showing no stimulation.

TABLE XIV (m)

*Bean roots* grown in distilled water with  $\text{CuSO}_4$ 

Cu p.p.m.	Elongation 48 hrs.	Height of tops
.1	2.9 mm.	87 mm.
.3	1.2	91
.5	.6	85

*Bean roots* grown in distilled water with  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ 

Cu p.p.m.	Elongation 48 hrs.	Height of tops
.1	2.9 mm.	98 mm.
.3	1.	88
.5	.6	84

Showing same behavior with  $\text{CuSO}_4$  and  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ .

TABLE XIV (n)

*Squash roots grown in distilled water with CuSO<sub>4</sub>*

Cu p.p.m.	Elongation 24 hrs.	Cu p.p.m.	Elongation 24 hrs.
check	3.6 mm.	.05	1.4 mm.
.01	2.8	.1	2.

*Squash roots grown in distilled water with Cu(OH)<sub>2</sub>.CuCO<sub>3</sub>*

Cu p.p.m.	Elongation 24 hrs.	Cu p.p.m.	Elongation 24 hrs.
check	3.6 mm.	.05	1.1 mm.
.01	3.8	.1	.4

Doubtful stimulation at .01 p.p.m.

TABLE XIV (o)

*Corn roots grown in distilled water with CuSO<sub>4</sub>*

Cu p.p.m.	Elongation 48 hrs	Cu p.p.m.	Elongation 48 hrs
check	8.7 mm.	.05	4.7 mm.
.01	10.9	.1	1.5

*Corn roots grown in distilled water with Cu(OH)<sub>2</sub>.CuCO<sub>3</sub>*

Cu p.p.m.	Elongation 48 hrs	Cu p.p.m.	Elongation 48 hrs
check	8.7 mm.	.05	3.3 mm.
.01	13.2	.1	2.

These cultures, while somewhat fragmentary, afford excellent indications of stimulating effects upon plant roots. Excluding squash, which is not satisfactory material to work with, corn and beans show consistent stimulations at very high dilutions. Measurements in all cases are averages of about ten observations.

TABLE XV

## SUMMARY OF STIMULATION EFFECTS

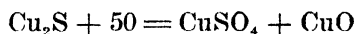
Experiment	Culture	Copper salt used	Character and strength in copper of solution producing stimulation	
			Well water <sup>a</sup>	Distilled water
<i>a</i>	Corn roots	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	at .01 p.p.m.	
<i>b</i>	Bean roots	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	.03	
<i>c</i>	Corn roots	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	.03	
<i>e</i>	Bean roots	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	.05	
<i>g</i>	Corn roots	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	.01-.1	
<i>h</i>	Corn roots	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>		none at .01 or above
<i>i</i>	Bean roots	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>	.05-.1	
<i>j</i>	Bean roots	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>		none at .01 or above
<i>o</i>	Corn roots	CuSO <sub>4</sub>		.01
<i>o</i>	Corn roots	Cu(OH) <sub>2</sub> .CuCO <sub>3</sub>		.01

Only at very high dilutions (one part of copper to from 10,000,000 to 100,000,000 of water) are accelerations of root growth observed. These occur with both corn and beans, in well water. In distilled water stimulation was observed only at the highest dilution—1:100,000,000. In well water stimulation was observed at from 1:100,000,000 to 1:10,000,000—consistently with the well known fact that in presence of other soluble salts the effects of copper are lessened.

## EFFECTS OF SOIL UPON TOXICITY OF COPPER SOLUTIONS

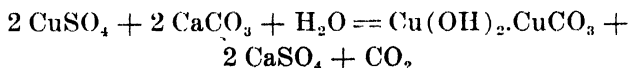
Of prime importance in connection with possible toxic effects of copper in soils are the various reactions (1) converting insoluble into soluble compounds, (2) reconverting these again into insoluble combinations, and (3) modifying the toxic effects of copper salts in solution.

As shown in the table of solubilities, both basic carbonate of copper and chrysocolla are soluble in carbon dioxide, forming solutions which in water cultures are highly toxic in character. Sulphides of copper are first oxidized to the sulphate, which is easily soluble:



For instance, 100 grams of chalcocite ore containing 3.2 per cent copper were shaken in a flask with 600 c.c. of water, frequently, during twenty-eight days. At the end of that time 500 c.c. of solution contained 0.0132 grams of copper.

Copper sulphate then reacts in the soil to form various insoluble compounds with consequent lessening of toxic action. With calcium carbonate the following represents a reaction which may occur:



For instance, two grams of precipitated carbonate of lime were added to an excess of ten grams of copper sulphate in one liter

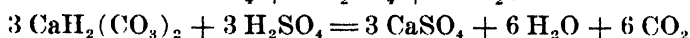
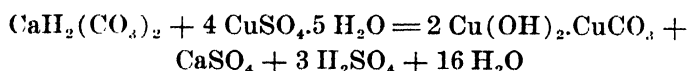
<sup>6</sup> "University of Arizona well water" contains 250 p.p.m. of soluble solids, mainly sodium sulphate.



of water, and digested with frequent shaking for over four months, the green precipitate being then filtered off, dried and analyzed for copper:

Weight of precipitate taken .....	100.00 mg.
Cu found .....	47.85
Theoretical Cu in basic carbonate .....	57.38

Indicating by the formula above a conversion to basic carbonate of copper of over 83 per cent of the solid carbonate of lime present. Bicarbonate of lime in solution also reacts with copper sulphate to form the basic carbonate



The silicates of the soil, also, and particularly those of zeolitic character, react readily with soluble copper compounds to form insoluble copper silicates. Organic matter likewise combines with large amounts of copper, to form compounds of indefinite or unknown composition. As a result of all these reactions, when soils are shaken up with solutions of copper salts the latter are withdrawn from solution in large amount. Under irrigation conditions, where waters containing minute amounts of copper are filtered through relatively large masses of soil, this action is nearly or quite complete.

Five large percolators were arranged with varying depths of

TABLE XVI  
PERCOLATION OF COPPER SOLUTIONS THROUGH SOILS

Solution used			Cu in solution, p.p.m.	Amount of percolate, c.c.	Copper in percolate, p.p.m.
Soil	Depth	Cu compound			
Sandy loam	1 in.	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ in $\text{CO}_2$ water	95	2000	none
Sandy loam	5 in.	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ in $\text{CO}_2$ water	95	1500	none
Sandy loam	9 in.	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ in $\text{CO}_2$ water	95	2000	none
Sandy loam	1 in.	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ in $\text{CO}_2$ water	56	2000	.85
Heavy clay containing .003% Cu	12 in.	$\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ in $\text{CO}_2$ water	8.5	600	
Heavy clay containing .003% Cu	12 in.	$\text{CuSO}_4 \cdot 5 \text{H}_2\text{O}$	254	150	7.3

soil resting on filter paper supported by a perforated porcelain plate. Two soils, heavy clay and sandy loam, were employed; and two copper solutions, sulphate and bicarbonate.

In nearly all cases copper as basic carbonate was entirely removed from solution in percolating through as little as a single inch of sandy loam. Although appreciable amounts of copper sulphate passed out of a soil, the latter in that case itself contained a very small percentage of copper. Inasmuch as soluble copper in irrigating waters must be present ordinarily as basic carbonate, its complete withdrawal by thin layers of soil is significant in connection with irrigated crops.

#### IRRIGATION EXPERIMENTS

A set of cultures was arranged to test the effects upon crop plants of solutions of basic copper carbonate so applied as to filter through the soil before reaching the plant roots. Six-inch

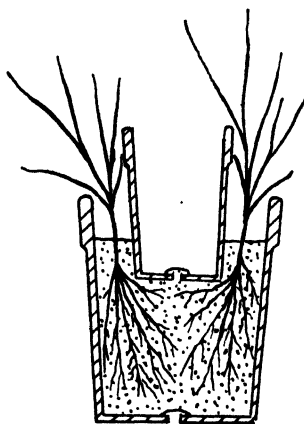


Fig. 5.—Diagram of pot culture irrigated through two-inch pot inside.

flower-pots were filled with sandy loam soil. In the middle of each of these pots a two-inch pot was half buried, and the plants experimented with were grown in the circles of soil between the large and small pots. These plants were irrigated by pouring the solution used into the small pot, through the bottom of which it passed, necessarily filtering through more or less soil before

reaching the plant roots. Radishes, beans, cantaloupes, cucumbers, lettuce, peas, beets, corn, berseem, avas, onions, barley, and wheat were employed; corn, barley, and wheat being especially successful under these conditions. All cultures were in pairs, one of each pair being irrigated with solutions of basic carbonate of copper in  $\text{CO}_2$ -water, and the check cultures with water only. In all other particulars—original strength of plants, exposure to light and air, and amount and time of watering—the conditions were identical.

These cultures were carried on in a greenhouse set aside for the purpose. The experiment was begun in November and ended the following March. The solutions of basic carbonate of copper employed contained from 0 to 55 p.p.m. of copper, averaging about 20 parts, which is from 7 to 670 times as much as has been observed in the waters of the Gila River from time to time.

TABLE XVII

CONDITION AT MATURITY OF CULTURES IRRIGATED WITH COPPER SOLUTIONS,  
AS COMPARED WITH THOSE IRRIGATED WITH WATER

		C, copper culture; W, check.	
		Tops	Roots
Radishes		C and W. The same in appearance and weight	The same, but in C roots were removed $\frac{1}{4}$ in. from inner pot hole
Beans	C greener	C and W. About the same	Equal; same number of nodules; very local effect of Cu at pot hole
Lettuce		C and W. About the same	The same except that in C roots were dead $\frac{1}{4} \times \frac{1}{2}$ in. under pot hole
Peas		C and W. Averaging the same	Both C and W having abundant nodules.
Beets			No apparent damage by Cu
Corn	Stimulated? C showing stronger	Weighing the same, but C appearing stronger	Fewer in soil under pot hole in C, otherwise equal
Berseem	C stimulated, earlier bloom	C more advanced in growth, but not so heavy	Equally developed, both showing strong nodule development.

		Tops	Roots
Avas		C and W. Same apparent growth	In C roots within $\frac{1}{2}$ in. of pot hole damaged. Both C and W show strong nodule develop- ment
Onions		C and W. Same general appearance	Very little local effect of Cu just under inner pot hole in C
Barley	C stimulated, ma- tured over twice as much grain	The same in weight, but C matured more grain	No roots in C for space of $1 \times \frac{1}{2}$ in. under inner pot hole
Wheat	C stimulated matured 20% more grain	Identical appear- ance, but C ma- tured more grain	In C no roots under in- ner pot hole for space of $1\frac{1}{2} \times \frac{1}{2}$ in.

In practically all cases a distinct but very local effect of copper solutions upon plant roots under the inner pot hole was observed. For a distance of a half-inch or less from the small pot hole exposed roots were dead or missing. The soil in this area was observed in two instances to contain 0.25 and 0.45 per cent copper, respectively. In one instance 80 per cent of the copper added was found in the 43 grams of soil just under the bottom of the little pot, showing the rapidity with which copper is removed from its solutions by filtration through the soil.

The tops of the cultures under consideration in no instance showed injury, but in certain cases were in a distinctly advanced condition. The amounts of copper contained in material derived from these cultures are as follows:

TABLE XVII (a)

## COPPER CONTENT OF PLANTS IRRIGATED WITH COPPER SOLUTIONS

Sample No.		Dry matter, grams	Cu grams	P.p.m. of Cu in dry material
3673	Wheat and barley tops grown in check			
3675	soil containing a trace (.0025 per cent) of copper .....	32.90	.000100	3.04
3672	Tops of beans, peas, corn, lettuce, car- rots, cucumbers and avas grown in check soil .....	133.00	.000350	2.60

Sample No.		Dry matter, grams	Cu grams	P.p.m. of Cu in dry material
3674	Tops of wheat and barley irrigated with water averaging 20 p.p.m. copper .....	30.70	.000400	13.00
3676	Tops of beans, berseem, peas, onions, lettuce, beets, radishes, corn, avas, barley, and wheat irrigated with water averaging 20 p.p.m. copper ....	27.20	.000751	27.60
3690	Roots of same (washed in 4 per cent HCl) .....	8.30	.000750	90.00

*In brief*, even when relatively large amounts of water containing excessive quantities of soluble copper were applied and the experiments so arranged that all of the copper remained in the limited volumes of soil employed, no general injury to the plants was observed, although apparently slight stimulation occurred in some cases. Prolonged irrigation with such solutions would be required to saturate the soil to a depth sufficient to seriously injure plants grown in it.

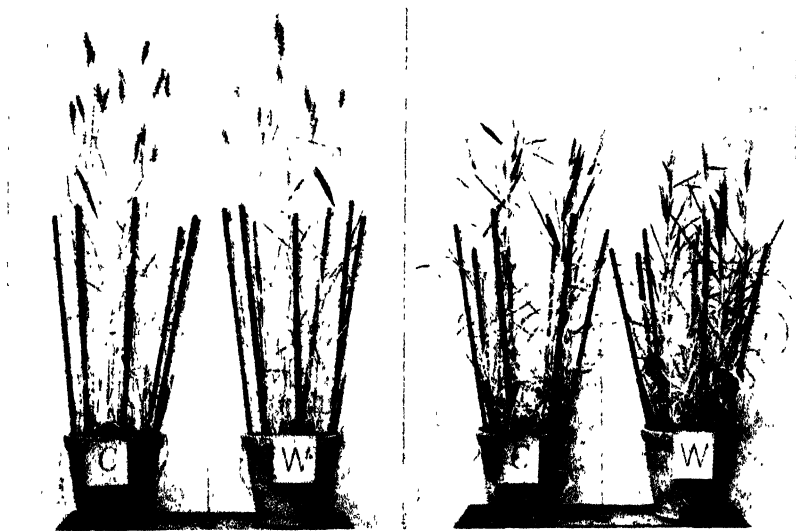


Fig. 6.—Wheat and barley irrigated (C) with copper solutions filtered through soil, and (W) with well water. Both show stimulated growth with copper.

## CULTURAL EXPERIMENTS

## POT CULTURES WITH TREATED SOILS

Pot cultures of corn, beans, and squash were also grown in soils containing copper in the form of precipitated carbonate ( $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ ), finely powdered (100-mesh) chalcocite or sulphide ore, and finely powdered chrysocolla or silicate ore. Large glazed stone jars containing thirty-eight pounds of soil were used. Effects on growth were observed and the copper content of tops and of root systems was determined. The following tabulations relate to the work done in this direction, the statement showing the copper content of corn, bean, and squash plants expressed in parts per million of copper in dry matter.

TABLE XVIII

## COPPER CARBONATE SERIES (1908), BEANS

Sample No.	Culture	Cu in soil, per cent	Appearance and height of plants of Normal	Dry matter, grams	Cu found, grams	Cu p.p.m. in	
						tops	roots
3944	Beans	Check*	39 in.	16.6	.00022	13	
4013	Beans	Check*		.72	.00033		453
3945	Beans	.01	38	17.2	.00027	16	
4014	Beans	.01		1.35	.00116		859
3946	Beans	.025	39	15.9	.00033	21	
4015	Beans	.025		1.21	.00115		950
Toxic effects begin at about .035% Cu in soil.							
			Stunted				
3947	Beans	.05	30	13.2	.00031	23	
4016	Beans	.05		1.09	.00148		1358
3948	Beans	.1	25	6.7	.00011	16	
4017	Beans	.1		1.44	.00212		1472
3949	Beans	.25	14	3.7	.00009	25	
4018	Beans	.25		1.35	.00243		1800
3950	Beans	.5	15	2.9	.0001	35	
4019	Beans			.87	.00147		1690
3951	Beans	1.	12	2.2	.00009	41	
4020	Beans	1.		.53	.00106		2000
3952	Beans	1.5	14	2.1	.00009	44	
4021	Beans	1.5		.5	.00115		2300

\* Containing traces of copper, .0025%.



Fig. 7.—Bean cultures grown in soils containing copper as precipitated carbonate, from none to 1.5 per cent Cu.

TABLE XIX

COPPER CARBONATE SERIES (1907), CORN

Sample No	Culture	Cu in soil, per cent	Dry matter, grams	Cu found, grams	Cu p.p.m. in	
					tops	roots
3870	Corn	Check*	45.2	.00020	1.40	
3885	Corn	Check*	8.8	.00035		40.00
3869	Corn	.01	175.9	.00115	6.50	
3868	Corn	.01	10.6	.00161		152.00
3865	Corn	.025	77.0	.00160	21.00	
3866	Corn	.025	9.2	.00670		728.00
3864	Corn	.05	47.7	.00103	22.00	
3867	Corn	.05	4.4	.00328		745.00
3863	Corn	.10	26.8	.00079	30.00	
3862	Corn	.15	9.8	.00046	47.00	
3861	Corn	.20	14.4	.00073	51.00	
3860	Corn	.30	4.6	.00110	239.00	

\* Containing traces of copper, .0025%.



Fig. 8.—Corn cultures grown in soils containing copper as precipitated carbonate, from none to .2 per cent Cu.

## COPPER CARBONATE SERIES (1908), CORN

Sample No.	Culture	Cu in soil, per cent	Appearance and height of plants	Dry matter, grams	Cu found, grams	Cu p.p.m. in roots
			Normal			
3992	Corn	Check*	43 in.	7.48	.00058	78.00
3993	Corn	.01	41	2.35	.00049	209.00
3994	Corn	.015	35	4.07	.00171	420.00
3995	Corn	.02	41	5.31	.00397	748.00
Toxic effects begin at about .023% Cu in soil.						
			Stunted			
3996	Corn	.025	33 in.	4.81	.00245	509.00
3997	Corn	.05	15	.31	.00023	742.00
3998	Corn	.10	22	3.62	.00651	1798.00
4000	Corn	.20	20	1.99	.00444	2231.00

\* Containing traces of copper, .0025%.

## COPPER CARBONATE SERIES (1908), SQUASH

Sample No.	Culture	Cu in soil, per cent	Appearance and height of plants	Dry matter, grams	Cu found, grams	Cu p.p.m. in	
			Normal			tops	roots
3937	Squash	Check*	16 in.	11.2	.00016	14.00	
3938	Squash	.01	16	6.3	.00023	36.00	
3939	Squash	.025	15	9.2	.00031	39.00	
4026	Squash	Chk., .01, and .025	.24	.00004			169.00

Toxic effects begin at about .035% Cu in soil.

			Blanched and stunted				
3940	Squash	.05	11 in.	3.7	.00017	46.00	
3941	Squash	.10	11	2.3	.00014	61.00	



Fig. 9.—Corn cultures grown in soils containing copper as sulphide (chalcocite), from none to 1. per cent Cu.



TABLE XX  
CHALCOCITE SERIES (1908)

Sample No.	Culture	Cu in soil, per cent	Appearance and height of plants	Dry matter, grams	Cu found, grams	Cu p.p.m. in	
						tops	roots
3979	Corn	Check*	36 in.	17.60	.00026	15.00	
3979c	Corn	Check*		4.62	.00027		58.00
3980	Corn	.01	33	11.60	.00011	10.00	
3980c	Corn	.01		2.94	.00023		78.00
3981	Corn	.02	38	19.40	.00021	11.00	
3981c	Corn	.02		6.14	.00114		186.00
3982	Corn	.03	35	17.90	.00028	16.00	
3982c	Corn	.03		6.99	.00176		252.00
3968	Corn	.05	45	51.70	.00065	13.00	
3978	Corn	.05		6.14	.00105		171.00

Toxic effects begin at about .08% Cu in soil.

			Stunted				
3983	Corn	.10	36 in.	14.00	.00031	22.00	
3983c	Corn	.10	yellow	6.08	.00625		1028.00
3984	Corn	.50	8 in.	3.20	.00040	125.00	
3984c	Corn	.50		.47	.00065		1383.00
3985	Corn	1.00	12	3.20	.00050	159.00	
3985c	Corn	1.00		.49	.00089		1816.00

\* Containing traces of copper, .0025%.

The cultures described in the foregoing tables indicate several interesting facts more or less applicable to field conditions.

(1) Precipitated carbonate of copper is shown to have a much more toxic effect upon corn than the finely pulverized ores of chalcocite or chrysocolla. With the precipitated carbonate 0.025 per cent in the soil was distinctly toxic, while with chalcocite and chrysocolla about 0.08 per cent was required to produce an equal effect. Inasmuch as all of these combinations of copper may occur in a soil subject to mining detritus, a mere determination of total copper in soils containing doubtfully toxic quantities cannot convey trustworthy information as to the injuriousness of the amounts present.

Moreover, since it has been shown that in the case of precipitated carbonate, and sulphate of copper, equivalent quantities of these salts in solution are equally toxic, it is probable that the greater toxicity of the carbonate is due to its greater solubility under soil conditions. It is, in fact, shown in table I, "Solubili-

TABLE XXI  
CHRYSOCELLA SERIES (1908)

Sample No.	Culture	Cu in soil, per cent	Appearance and height of plants	Dry matter, grams	Cu found, grams	Cu p.p.m. in	
						tops	roots
4003	Corn	Check*	Normal 32 in.	25.60	.00025	10.00	
4003c	Corn	Check*		6.46	.00012		19.00
4004	Corn	.05	33	23.50	.00026	11.00	
4004c	Corn	.05		6.70	.00062		93.00
Toxic effects begin at about .08% Cu in soil.							
			Dwarfed				
4005	Corn	.10	30 in.	17.90	.00024	13.00	
4005c	Corn	.10	striped	5.82	.00094		162.00
4006	Corn	.10	28 in.	10.50	.00017	16.00	
4006c	Corn	1.00	yellow	4.29	.00233		543.00

\* Containing traces of copper, .0025%.



Fig. 10.—Corn cultures grown in soils containing copper as silicate (chryso-colla), from none to 1. per cent Cu.

ties of Copper Compounds," that precipitated copper carbonate is soluble to the extent of 1.5 parts in 1,000,000 of water, while copper sulphide is soluble to the extent of 0.09 parts of copper in 1,000,000 of water. It is most probable, also, that the finely divided condition of the precipitated carbonate is more favorable to solution, and also to reaction with the acids of plant roots.

(2) Corn is seen to be distinctly more sensitive to the carbonate of copper than either beans or squash. With corn, toxic effects appear with 0.02 per cent of copper in the soil, while with beans and squash these toxic effects do not appear until 0.035 per cent of copper in the soil is reached. As is suggested

in the following pages, the physical constitution of root systems may account in part for varying degrees of sensitiveness to copper compounds.

The presence of copper in tops and roots of check is due to 0.0025 per cent of copper in the soil which was supposed originally to be free from this element.

#### POT CULTURES WITH FIELD SOILS

Two field soils containing copper from irrigating waters were tested in pot culture with reference to toxic effects and



Fig. 11.—Pot cultures of corn in field soils containing tailings. No. 3887, .027% Cu; no. 3888, .047% Cu; and no copper. Cultures in field soils are slightly affected.

copper content of root systems. The soils employed were from a field showing varying effects of accumulations of tailings, immediately southeast of Safford:

Sample	Cu in soil, per cent
3887 Sandy loam, surface 12 in. of soil recently put under irrigation .....	.027
3888 Heavy clay (tailings) mixed with sandy loam, surface 12 in., long under irrigation, much tailings .....	.047

In these two soils, differing mainly through the addition of tailings to No. 3888, cultures of corn, beans, and squash were made, and examined for copper with the following results:

TABLE XXII  
CULTURES IN TAILINGS SOILS

No.	Pot culture	Condition	Cu in soil, per cent	Cu p.p.m. in	
				tops	roots
3887	Corn in sandy loam	Distinctly striped	.027		453.00
3888	Corn in sandy loam and tailings	Less distinctly striped	.047		163.00
3887	Beans in sandy loam	Normal appearance	.027	28.00	1523.00
3888	Beans in sandy loam and tailings	Normal appearance	.047	19.00	703.00
3887	Squash in sandy loam	Yellow and stunted	.027	73.00	
3888	Squash in sandy loam and tailings	Normal appearance	.047	45.00	



Fig. 12.—Showing effects of copper modified by tilth of soil. Strong growth, lumpy mixture; weak growth, thoroughly mixed.

Bean cultures appeared little affected by copper in either No. 3887 or No. 3888; but squash was distinctly damaged in No. 3887, being yellow and stunted. The leaves of both cultures of corn were paler than those of the check, but in soil No. 3887, containing less copper, the leaves of corn were more distinctly striped than in No. 3888. This is probably due to the sandy character of No. 3887 with consequently decreased adsorptive action upon copper salts. Lumpiness in the heavier soil might also account for a lessened toxic action, as indicated by an experiment in which 0.1 per cent of copper in the form of pre-

cipitated carbonate was mixed (1) intimately and (2) in lumpy condition. Results were as follows:

Sample No.	Cu as pptd. carbonate, per cent	Condition	Cu p.p.m. in roots
3998c	0.1 well mixed	22 in. high, much blanched	1798.00
3999c	0.1 lumpy	28 in. high, mostly green	457.00

In these instances it may be noted that toxic effects are associated with higher copper content of roots of plants, rather than with copper content of soils employed.

As in other cultures it is observed that beans, though carrying a higher copper content than corn, show less toxic effects—a fact possibly to be explained by the higher protein content of the plant with a consequently greater capacity for absorption of copper before toxic effects appear.

#### POT AND PLOT CULTURES

In order to carry experimental cultures further towards field conditions, cultures of wheat and corn in small plots of sandy loam garden soil,  $2\frac{1}{2} \times 18$  feet, were grown, copper in the form of finely powdered sulphate having been thoroughly spaded in four times to a depth of nine inches in the amounts shown in table XXIII. The roots of these cultures were harvested and examined as usual for copper.

TABLE XXIII

CORN GROWN IN GARDEN PLOTS CONTAINING CU APPLIED AS  $\text{CuSO}_4$ , (1914)

Sample No.	Cu added, per cent	Condition of leaves	Dry matter, grams	Cu found, grams	Cu p.p.m. roots
5858a	none	Solid green	11.0	.00015	14.00*
Toxic effects begin at about .008% Cu in soil.					
5859a	.01	Distinctly yellow striped	11.6	.00117	101.00
5860a	.025	Distinctly yellow striped	8.6	.00211	246.00
5861a	.05	Distinctly yellow striped	7.3	.00215	296.00
5862a	.10	Strongly yellow striped	4.3	.00300	698.00
5863a	none		6.2	.00013	21.00*

\* Probably resulting from roots spreading to copper soils.

TABLE XXIV

WHEAT GROWN IN GARDEN PLOTS CONTAINING CU AS  $\text{CuSO}_4$  (1914)

Sample No.	Cu added, per cent	Condition of leaves	Dry matter, grams	Cu found, grams	Cu p.p.m. in roots
5648a	none	29 in. high; good	4.46	.00012	27.00
5649a	.01	29 in. high; good	3.16	.00190	601.00
Toxic effects begin at about .02% Cu in soil.					
5650a	.025	25-27 in. high; affected	3.23	.00260	805.00
5651a	.05	23 in. high; severely affected	1.90	.00330	1737.00
5652a	.10	20 in. high; very severely affected	1.33	.00200	1504.00

TABLE XXV

WHEAT GROWN IN POTS TO CHECK PLOTS CONTAINING CU AS  $\text{CuSO}_4$  (1914)

Sample No.	Cu added, per cent	Condition of leaves	Dry matter, grams	Cu found, grams	Cu p.p.m. in roots
5672a	.0025	Green; 27 in. high	1.51	.00007	46.00
Toxic effects begin at about .005% Cu in soil.					
5673a	.01	Yellowish; 23 in. high	1.96	.00035	179.00
5674a	.025	Yellow and stunted; 17 in. high	.84	.00030	357.00
5675a	.05	Yellow and stunted; 12 in. high	.52	.00031	593.00
5676a	.10	Yellow and stunted; 4-12 in. high	.30	.00044	1476.00

The corn series contains much smaller proportions of copper in the roots than either of the wheat series, a fact explained in part by the coarser roots of corn, which therefore have less absorptive surface in proportion to their weight. Wheat roots grown in plots show much more copper than pot samples, although the copper is much more toxic to the plants in pots than in plots, a contradiction not easily understood unless it be that other less favorable conditions of growth in pots were responsible for the backward condition of the plants.

#### FIELD SAMPLES OF SOILS AND VEGETATION

In order to relate, if possible, the experimental work detailed on previous pages to samples of field material, roots of barley, wheat, oats and corn, were collected in the district studied and the amounts of copper in them determined. The samples of bar-

TABLE XXVI

COPPER IN SOILS, AND IN ROOTS OF PLANTS GROWN IN FIELD SOILS CONTAINING MINING DETRITUS, NEAR SOLOMONVILLE AND SAFFORD (1914)

		Dry matter, grams	Cu found, grams	Cu p.p.m. in	
				tops	roots
Jan. 3, 1909					
4008a	Barley tops selected for toxic effects from tailings soil (Wm. Gillespie), Solomonville, under Montezuma Canal .....	13.90	.00061	43.80	
4008b	Barley roots, ditto .....	2.70	.00160		592.50
4009a	Oat tops selected for toxic effects from field one mile west of Solomonville, under Montezuma Canal	28.30	.00121	42.70	
4009b	Oat roots, ditto .....	2.55	.00025		98.00
March, 1914					
		Dry matter, grams	Cu found, grams	Cu p.p.m.	
				yellow	green
5544a	Barley roots, yellow plants ..	3.78	.00037	98	
5545a	Barley roots, green plants ....	2.33	.00290		124
5546a	Barley roots, yellow plants ..	3.41	lost	.....	
5547a	Barley roots, green plants ....	3.80	.00047		123
5548a	Wheat roots, yellow plants ....	1.40	.00044	314	
5549a	Wheat roots, green plants .....	1.25	.00048		382
5550a	Barley roots, less green plants .....	2.17	.00077	354	
5551a	Barley roots, stronger plants	3.34	.00110		329
5552a	Oat roots, yellow plants .....	1.67	.00066	394	
5553a	Oat roots, green plants .....	1.77	.00030		169
5554a	Barley roots, yellow plants ..	1.38	.00057	411	
5555a	Barley roots, green plants ....	1.42	.00041		289
Average .....				314	236
4010	Corn roots (1908) in tailings soil, Solomonville .....	16.12	.00097		60
November, 1914					
5841a	Corn roots, tailings 9 in. deep	10.18	.00021		21
5842a	Corn roots, tailings 8 in. deep	11.08	.00038		34
5843a	Corn roots, old tailings .....	10.01	.00038		38
5844a	Corn roots, tailings 6-12 in. deep .....	21.48	.00039		18
5845a	Corn roots, old tailings .....	16.24	.00068		42
5846a	Corn roots, old tailings .....	5.02	.00034		68
5847a	Corn roots, old tailings .....	14.42	.00104		72
Average .....				42	

ley, wheat and oats were collected in sets of two in a place. One of each set was green, healthy growth, the other more or less yellow and unthrifty in appearance. The object of this method of sampling in soils found to contain small amounts of copper was, if possible, to relate unthrifty appearance of plants examined to copper found in roots and surrounding soil. Table 26 (p. 441) contains the results of the determinations made.

As may be expected under field conditions, which are more complex and variable than those of plot or pot cultures, these data are considerably contradictory. Roots of yellow barley, wheat and oat plants, for instance, in 5544a and 5548a contain less copper than roots of strong green plants grown alongside; although the average copper content (314 parts) of yellow and more or less unthrifty plants is seen to be greater than in green plants alongside (236 parts). So far as observed, the larger percentages of copper found in soils shaken from roots of the plants are always associated with yellow plants. The average copper content of soils from roots of yellow plants is 0.048 per cent, while that from green plants is 0.023 per cent. These observations indicate that in a general way the larger amounts of copper found in these field soils are associated with larger amounts of copper in root systems and with yellow color in young plants.\* The percentages of copper observed in the soil, ranging up to 0.073 per cent in one instance, is surprisingly high, but toxic effects must be qualified by the character of the compounds, soluble salts in the soil, and other factors noted on preceding pages.

Yellowness of foliage also may be due to other causes than copper. Among these are: (1) too much water, as in low places; (2) alkali accumulations; (3) cold weather; (4) too much nitrogen in improper form, as in some old barnyards; (5) too little available nitrogen, as on new ground; (6) shade, and (7) insect pests and plant diseases. Malnutrition from any cause, in fact, usually expresses itself in the yellow or striped appearance of the leaves of these crop plants. Such appearance, therefore, cannot be attributed to copper present in the soil, without exclusion of other causes and sufficient confirmatory evidence.

As in the case of plot and pot cultures, corn roots are ob-



served to contain much less copper than other grain roots grown in similar soils, a fact to be attributed to the coarse character of field samples of corn roots.

### USE OF COPPER SULPHATE TO KILL MOSS IN IRRIGATING DITCHES

Clear irrigating water supplies, such as are derived from seepage and from wells, quickly become choked with mosses and algae in warm weather, entailing loss of water and expensive ditch cleaning. In order to test the application of copper to a running stream for the purpose of killing the growth of aquatic plants, an experiment was conducted, in October, 1906, upon the Flowing Wells ditch near Tucson, which at the time contained abundant aquatic growth.

A barrel of copper sulphate solution was prepared and placed at the head of the ditch. By means of a small outlet controlled by a stopcock, fifteen pounds per hour of  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$  were added to the ditch flow, this amount being in the proportion of 1 part of copper to 100,000 of water. Most of the copper was immediately precipitated by the bicarbonate of lime present in the water; still more probably combined in insoluble form with the soil along the ditch; while the remainder acted with toxic effect upon the sensitive algae and the less sensitive mosses (*Potamogetons*) growing in the water. A short distance below the barrel, where algae and mosses, after twenty-five hours' exposure to copper, were brown and dead and breaking away from their points of attachment. .84 parts of copper in 1,000,000 of water remained in solution. Three miles below the barrel, where the mosses and algae were still plainly affected, traces only of dissolved copper were perceptible. A renewal of copper from point to point would therefore have been necessary in treating a long ditch by this method, which, however, proved too costly for adoption in the instance mentioned.<sup>7</sup>

It is of interest in this connection to note that in the early days of irrigation on the Gila River, mosses grew in such abundance in the clearer waters obtained from the river at that time,

<sup>7</sup> See Bibliography, p. 488, reference 32.

that considerable labor was required to keep the ditches clean. These mosses have now entirely disappeared from the upper canals, due in part to the turbid waters in which they will not grow, and in part, perhaps, to the dissolved copper from the mines.

## PHYSIOLOGICAL OBSERVATIONS ON TOXIC EFFECTS OF COPPER SALTS

### QUANTITATIVE WORK

Citrus seedlings placed in copper sulphate solutions containing from 2.5 to 100 parts of copper in 1,000,000 of distilled water wilted in forty-eight hours, thus showing effects of toxicity. Root tips then all turned red with  $K_4FeCy_{16}$ . Red root-tips sectioned showed under low power red cells under bark and around center. Citrus, cucumber and bitter melilot roots grown in 10:1,000,000 copper solution all gave violet reaction with KOH, less delicate but more distinctive than  $K_4FeCy_{16}$ , since the purple biuret test indicates both copper and protein.

Cultures of wheat, peas, corn, beans, and other plants grown in soils containing from 0.005 to 0.1 per cent of copper in soil, gave only very doubtful root-tip reactions with  $K_4FeCy_{16}$ , although showing evident injury, especially in 0.1 per cent culture. There is an essential difference between water-culture roots *placed in* copper solutions and roots *grown in* soil. The first are killed by excess of copper salts contained; the second are yet living and growing resistantly in the soil.

A 0.1 per cent copper culture of corn, wheat, beans and cucumbers was washed out from the soil and gave superficial red coloration with  $K_4FeCy_{16}$ , but not internal. Living tissue is evidently inconsistent with sufficient amounts of copper to give a plain internal test. Therefore, the small amounts of copper known to be in poisoned but living root systems must be disseminated. It is, therefore, of interest to know the copper-protein ratio in poisoned but living root systems, such a ratio being more significant than the ratio of copper to the whole mass of root systems, which includes various proximate principles not concerned in copper fixation.

Two assembled samples of corn, radish, wheat, vetch and peas grown in soils containing 0.005 per cent and 0.05 per cent of copper were, therefore, very thoroughly washed out, copper determined, and nitrogen determined  $\times 6\frac{1}{4}$  for protein. The amount of copper required to saturate vegetable protein was assumed at 11.7 per cent (14.655 per cent CuO)—the average of figures given in Mann's *Chemistry of the Proteids*, page 305.

(1) Roots grown in .005% Cu in soil .....	.4739000 gm.
Cu .0105% .....	.0000498
N. 2.26% = Protein 14.125% .....	.0669400
Cu required for saturation of protein	
.06694 gm. $\times$ 11.7% = .007832 gm. Cu for saturation	
Per cent saturated = $\frac{.0000498}{.007832}$ = .637%	
(2) Roots grown in .05% Cu in soil .....	.3561000 gm.
Cu .0322% .....	.0001147
N. 2.76% = Protein 17.25% .....	.0614900
Cu required for saturation of protein	
.06149 gm. $\times$ 11.7% = .007194 gm. Cu for saturation	
Per cent saturated = $\frac{.0001147}{.007194}$ = 1.594%	

Summary:	Cu p p m dry roots	Per cent saturation of protein with copper
(1) .005% Cu in soil .....	105	0.636
(2) .05% Cu in soil .....	322	1.594
Ratio (1) to (2) .. ...	3.07	2.51

*In brief*, 10 times as much copper in the soil resulted in 3 times as much copper in the entire root systems and 2.5 times as much in the protein of these root systems. This latter increase, however, is responsible for an increase in damage from almost nothing to very severe.

Further observations on the copper-protein saturation figure in roots grown in soil containing copper, were made on wheat and Canada peas, planted in pots containing soil mixed with varying percentages of copper in the form of precipitated basic carbonate. The pots contained 102 pounds of sandy loam, and were irrigated in a uniform manner from time to time as water was needed. Plantings were made January 3, 1916, and roots harvested May 15.

TABLE XXVII

OBSERVATIONS ON THE SATURATION WITH COPPER OF PROTEIN IN ROOTS  
GROWN IN SOIL TREATED WITH  $\text{Cu}(\text{OH})_2 \cdot \text{CuCO}_3$ 

Lab. No.	Copper in soil, per cent	Material	Weight of sample, grams	Weight of Cu in sample, grams	Protein in sample, grams	Cu required to saturate protein, grams, factor 11.7%	Percentage of saturation of protein with Cu
6396	.005	Wheat roots	2.4726	.00023	.1069	.0125	1.84
6397	.02	Wheat roots	4.0122	.00068	.1660	.0194	3.50
6398	.06	Wheat roots	2.2236	.00068	.1231	.0144	4.70
6399	.10	Wheat roots	2.4658	.00037	.1401	.0164	2.25
6401	.005	Canada pea roots	2.6275	.00085	.3936	.0461	1.85
6402	.02	Canada pea roots	2.3844	.00093	.3684	.0431	2.16
6403	.06	Canada pea roots	2.0056	.00053	.2657	.0311	1.70
6404	.10	Canada pea roots	2.2708	.00093	.3747	.0438	2.12

While the figures on saturation in the last column of the table vary without reference to the amount of copper in the soil and the degree of injury observed in the roots, yet they all show a very low ratio of copper found to copper required for saturation of protein present.

In both wheat and peas, injury was first shown at 0.02 per cent of copper in soil, increasing greatly with higher percentages. This injury, showing as a characteristic crinkly condition, is best seen in wheat and corn and has been observed in wheat roots grown in a soil containing as little as 0.017 per cent of copper.

A further quantitative study of copper effects on root systems was carried out in water culture with corn, wheat, and Canada peas. Paraffin (parowax) disks one-third of an inch thick and nine inches in diameter were employed, perforated with holes of suitable diameter by means of steel cork borers. These disks were supported on paraffin posts two and one-half inches high in four-quart deep graniteware pans containing the water culture solutions which were used. After soaking, the germinating seeds were planted in paraffin disks of suitable perforation and nutrient solution was then poured up to level of contact with seeds. At first tap-water was used; then a nutrient solution made up as follows:

KNO <sub>3</sub> .....	1.0 gm.
MgSO <sub>4</sub> .....	.05
NaCl .....	.5
CaSO <sub>4</sub> .....	.5
FeCl <sub>3</sub> .....	.04
Tap-water .....	1.0 liter

No phosphate was included because of its precipitating action on copper salts. After the cultures were about four weeks old they were changed to nutrient solutions containing small amounts of copper which was gradually increased from one to ten parts per million of solution. The solution was neutralized with normal H<sub>2</sub>SO<sub>4</sub> (methyl orange indicator) to prevent precipitation of copper by dissolved carbonates. Following is a summary of the history of the cultures, each of which was increased to include several hundred plants:

#### WHEAT

- Dec. 16 Planted in tap-water.
- Dec. 24 Transferred to nutrient solution, one-third strength.
- Jan. 8 Changed to nutrient solution, two-thirds strength.
- Jan. 14 To nutrient solution, full strength.
- Jan. 17 To nutrient solution containing 1 part Cu per million.
- Jan. 21 To nutrient solution containing 2 parts Cu per million.
- Jan. 25 To nutrient solution containing 6 parts Cu per million.
- Jan. 28 To nutrient solution containing 10 parts Cu per million.
- Feb. 9 Experiment terminated.

A faint biuret test appeared after addition of 6 p.p.m. Cu. Also distinct K<sub>4</sub>FeCy<sub>6</sub> test. Roots did not become flaccid, but the tops of cultures were dying back and prostrated markedly in comparison with roots of control culture.

#### CANADA PEAS

- Dec. 20 Planted in tap-water.
- Dec. 30 Transferred to fresh tap-water.
- Jan. 6 Transferred to fresh tap-water.
- Jan. 18 Transferred to nutrient solution.
- Jan. 20 Changed to nutrient solution containing 1 part Cu per million.
- Jan. 21 To nutrient solution containing 2 parts Cu per million.
- Jan. 25 To nutrient solution containing 6 parts Cu per million.
- Jan. 28 To nutrient solution containing 10 parts Cu per million.
- Feb. 6-8 Experiment terminated.

A faint biuret test appeared after addition of 6 p.p.m. Cu. Distinct K<sub>4</sub>FeCy<sub>6</sub> test in root tips at end of experiment. Roots not flaccid, but plants distinctly affected and tops dying back more than those of control culture.

## CORN

- Dec. 22 Planted in tap-water.  
 Jan. 8 Changed to nutrient solution, one-third strength.  
 Jan. 19 To nutrient solution containing 1 part Cu per million.  
 Jan. 21 To nutrient solution containing 2 parts Cu per million.  
 Jan. 25 To nutrient solution containing 6 parts Cu per million.  
 Jan. 28 To nutrient solution containing 10 parts Cu per million.  
 Feb. 9 Experiment terminated.

Giving distinct, faint biuret test after addition of 6 p.p.m. Cu; also  $K_4FeCy_6$  test at end of experiment. Roots not flaccid at end of experiment, but tops of cultures about half dead, while tops of control culture were still in good condition.

These cultures, as shown by the notes, were exposed to copper solutions—wheat twenty-three days, peas eighteen days, corn twenty-one days. At the end of the experiment roots were not flaccid, but very faint biuret and distinct ferrocyanide tests were observed. In all cases top growth was affected, corn most, wheat next, and peas least. This material, as indicated above, is poisoned only just enough to show reactions in root tips, although tops are distinctly affected. It, therefore, represents minimum rather than maximum toxic conditions. Material was harvested and analyzed to show copper and nitrogen ratios; and by estimating the number of root tips in samples of corn, peas, and wheat the amount of copper per root tip, required to show faint tests, was found.

TABLE XXVIII

QUANTITATIVE DETERMINATIONS ON WATER CULTURES SHOWING SLIGHT  
TOXIC EFFECTS

CORN				
No.	Sample	Dry matter, grams	Cu found, grams	Cu p.p.m. in dry matter
6321	265 tops	17.695	.00048	27.10
6320 } 6323 }	Coarse roots	2.4277	.00022	91.00
6319	1100 root tips	.77	.00042	545.50
Amount of copper per root tip associated with slight toxic effects, .00042 ÷ 1100 = .00000382 gm.				

## PEAS

No.	Sample	Dry matter, grams	Cu found, grams	Cu p.p.m. in dry matter
6327	250 tops	7.0850	.00012	16.90
6325	Coarse roots	1.2827	.00180	1400.00
6326	Fine roots	.8393	.00141	1680.00
6324	5500 root tips	.4462	.00153	3428.00

Amount of copper per root tip required to show slight toxic effects,  
 $.00153 \div 5500 = .000000278$  gm.

Total roots examined for nitrogen ..... 4.41730 gms.

Albuminoids in roots (Alb. N.  $\times 6\frac{1}{2}$ ) ..... .83929

Copper required to saturate albuminoids (factor 11.7%) .09819

Total Cu found ..... .00784

Saturation .....  
 $.00784 \div .09819 = 7.99\%$

## WHEAT

No.	Sample	Dry matter, grams	Cu found, grams	Cu p.p.m. in dry matter
6332	530 tops	12.7	.00165	129.90
6331	Roots	.6902	.00020	297.00
6330	16000 root tips	.3664	.00103	2811.00

Amount of copper per root tip associated with slight toxic effects,  
 $.00103 \div 16000 = .000000064$  gm.

Total roots examined for nitrogen ..... 2.9223 gms.

Albuminoids in roots (Alb. N.  $\times 6\frac{1}{2}$ ) ..... .30794

Copper required to saturate albuminoids (factor 11.7%) .03603

Total Cu found ..... .001789

Saturation .....  
 $.001789 \div .03603 = 4.96\%$

These figures show, as usual, relatively small amounts of copper in tops of plants, with large amounts in roots, increasing from coarser to finer portions, until in the root tips corn contains 545, peas 3428, and wheat 2811 parts per million of copper in dry matter. For peas and wheat these are the largest proportions of copper thus far observed in any plant samples.

When the total amount of copper found in each sample is divided by the number of root tips employed, an extraordinarily small amount of copper is found necessary to bring about toxic effects. For instance

One corn root tip (terminal 3 cm.) required....	.000000382 gms. Cu
One pea root tip (terminal 1 cm.) required.....	.000000278 gms. Cu
One wheat root tip (terminal 1 cm.) required	.000000064 gms. Cu

Moreover, the extent to which albuminoids in affected roots are saturated with copper—only 7.99 per cent for peas and 4.96 per cent for wheat—indicates a maximum effectiveness upon roots of small amounts of the metal.

#### REACTIONS OF COPPER WITH GROWING POINTS

Corn seedlings fifteen days old were fixed with cotton in tall 50-c.c. graduated Nessler tubes containing different strengths of copper sulphate in pure distilled water. The strengths of solution employed were 20, 10, 5, 2.5, and 1.25 p.p.m. There was a check culture with no copper. After three days, in all cases except the check, the roots were flaccid, showing contraction on graduations and giving biuret and ferrocyanide tests, increasing in strength from weaker to stronger concentration.

An experiment with pea seedlings gave similar results, but when the quantity of pea roots was increased and weak solutions, 2.5 and 1.25 p.p.m., were employed in small quantities (20 c.c.), the tests became much fainter.

Severed roots of corn, also, were observed to give as good tests as roots of living plants. A large number (seventy) of severed root tips placed in a small quantity (20 c.c.) of weak solution (5 p.p.m.) gave only a faint ferrocyanide test. These observations indicate that the concentration of copper in growing points is due to ionic dissociation and migration through the semi-permeable membranes of the root systems,<sup>8</sup> rather than to transpiration. The fainter test for copper in large quantities of root material indicates lessened toxicity of dilute solutions of copper in presence of excess of root materials.

Mature wheat, corn and pea plants in nutrient solutions, but not growing actively, were treated with gradually increasing amounts of copper from January 21 to February 2, as follows:

#### WHEAT, CORN, AND PEA PLANTS, THIRTY-SEVEN DAYS OLD, TREATED WITH COPPER IN NUTRIENT SOLUTION

Jan. 21-25; nutrient sol. w. 2 p.p.m. Cu.

Jan. 25-28; nutrient sol. w. 4 p.p.m. Cu.

Jan. 28 to Feb. 5, nutrient sol. w. 10 p.p.m. Cu.

<sup>8</sup> See Bibliography, p. 488, references 35, 36, 37, 38, 39, 40, 41, 42, 43, 44.



Feb. 5; very faint biuret test for copper, distinct ferrocyanide test.

Corn, forty-eight days old in 50 p.p.m. Cu sol., two days, gave faint biuret and ferrocyanide tests.

Corn, forty-eight days old in 500 p.p.m. Cu sol., two days, gave faint tests for copper.

From these observations it is evident that the nearly negative results shown are due either to nutrient salts present or to the older and therefore more quiescent material employed. To settle this question, the following experiments were made:

(1) Young (ten days) wheat and corn plants were placed in copper solution in distilled water and in nutrient solutions and observed after twenty and forty hours, as follows:

2.5 p.p.m. Cu, distilled water—20 hours

10 days old: Young wheat; flaccid?; strong biuret test; strong  $K_4FeCy_6$  test

60 days old: Old wheat; not flaccid; no strong biuret test; distinct  $K_4FeCy_6$  test

10 days old: Young corn; flaccid; strong biuret test; strong  $K_4FeCy_6$  test

60 days old: Old corn; flaccid; no biuret test; old tips, faint  $K_4FeCy_6$  test  
young tips, strong  $K_4FeCy_6$  test

10 p.p.m. Cu, distilled water—20 hours

10 days old: Young wheat; flaccid?; strong biuret test; strong  $K_4FeCy_6$  test

60 days old: Old wheat; not flaccid; faint biuret test; distinct  $K_4FeCy_6$  test

10 days old: Young corn; flaccid; strong biuret test; strong  $K_4FeCy_6$  test

60 days old: Old corn; flaccid?; distinct biuret test; strong  $K_4FeCy_6$  test

40 p.p.m. Cu, distilled water—20 hours

10 days old: Young wheat; flaccid; strong biuret test; strong  $K_4FeCy_6$  test

60 days old: Old wheat; flaccid?; faint biuret test; distinct  $K_4FeCy_6$  test

10 days old: Young corn; flaccid; strong biuret test; very strong  $K_4FeCy_6$  test

60 days old: Old corn; flaccid; strong biuret test; strong  $K_4FeCy_6$  test

The above results indicate that old roots of corn and wheat are more resistant to the penetration of copper than are the young roots. This is shown by less flaccidity in the weaker solutions and by the fainter tests observed. A second series with greater strengths (5, 20, and 100 p.p.m.) and longer exposure (forty-five hours) showed distinctly less differentiation than in the case of the series above given in detail. This is to be expected, inasmuch as stronger solutions must overcome resistance of roots exposed to them more quickly, and the longer time employed would likewise tend to overcome differences existing in the first few hours of the experiment.

(2) Young and old wheat and corn roots were placed in 10 p.p.m. Cu in distilled water and 10 p.p.m. Cu in nutrient solution, with the following results:

10 p.p.m. Cu, distilled water—20 hours

10 days old: Young wheat; flaccid†; strong biuret test; strong  $K_4FeCy_6$  test

60 days old: Old wheat; not flaccid; faint biuret test; distinct  $K_4FeCy_6$  test

10 days old: Young corn; flaccid; strong biuret test; strong  $K_4FeCy_6$  test

60 days old: Old corn; flaccid; distinct biuret test; strong  $K_4FeCy_6$  test

10 p.p.m. Cu, neutralized nutrient solution—20 hours

10 days old: Young wheat; not flaccid; doubtful biuret test; faint  $K_4FeCy_6$  test

60 days old: Old wheat; not flaccid; none or doubtful biuret test; faint  $K_4FeCy_6$  test

10 days old: Young corn; not flaccid; faint biuret test; distinct  $K_4FeCy_6$  test

60 days old: Old corn; not flaccid; distinct biuret test; distinct  $K_4FeCy_6$  test

This shows very distinctly the prevention of toxic action upon plant roots through the protective action of other solids in solution, as already observed in water cultures by measurements of root growth. It is noteworthy in this connection that corn roots generally seem to be more sensitive to the action of copper salts than the roots of wheat or peas.

In order to examine still further into the relative resistance of old and young root systems to copper salts, a solution of 5 p.p.m. Cu in distilled water was used, the time being varied from twenty to two hundred hours. The results of these observations indicate that, with wheat and corn roots, the penetration of copper is distinctly more rapid in young than in old material. Peas did not give clear results.

It appears from these observations, first, that the accumulation of copper in plant roots is distinctly due to the migration of dissociated ions into the root systems, where they are fixed by protoplasm, in which combination they are identified by means of the biuret test. Second, the presence of nutrient salts very distinctly lessens the effect of a 10 p.p.m. copper solution upon sensitive young growing plant roots. Third, old quiescent plant roots developed in a nutrient solution are distinctly less sensitive to copper salts than young roots which are still actively growing.

The slow development of biuret tests for copper in such material after sufficient exposure to copper solutions, indicates the presence of protoplasm.

It is possible that the same observations may apply to other poisons, metallic or otherwise, brought into contact with absorp-

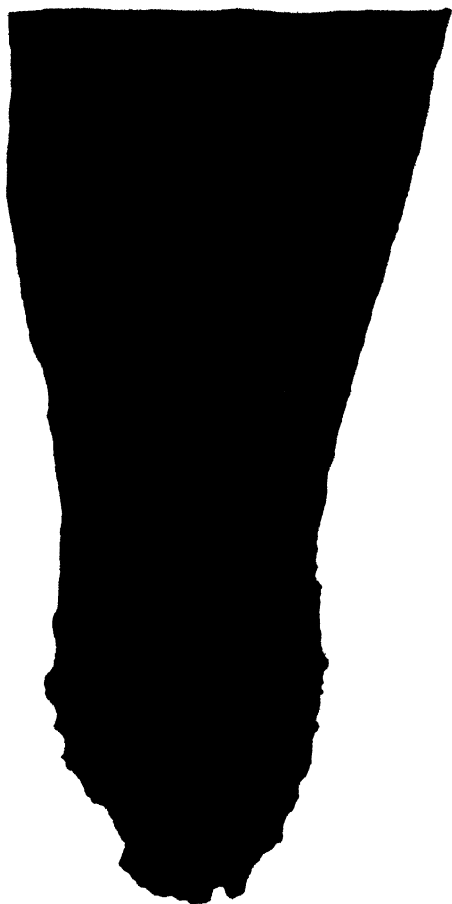


Fig. 13.—Photomicrograph of root tip of corn grown in water culture and poisoned by 1:200,000 of Cu in solution. The copper is shown as red copper ferrocyanide, which appears black in the photomicrograph. The irregular inner black line shows the penetration of the copper and also indicates sharply the differences in permeability of adjacent cells, some of which are penetrated before others. ( $\times 80$  diam.) (Photo by J. T. Barrett.)

tive root systems in the soil. Not only this, but it may be true that nutrient salts, as well, will be found more actively absorbed by younger and more sensitive root systems than by older ones, or by root systems which for any reason have become quiescent. This would suggest the possibility of choosing to advantage the proper time for applying substances, either to avoid injury or, as in the case of fertilizers, to secure maximum benefit from them.

#### VARYING RESISTANCE OF INDIVIDUAL CELLS TO COPPER

Not only do old and young roots vary as to toxic effects upon them of copper, but different degrees of resistance between individual cells in the same root and even in the same chain of cells, is clearly shown in the photomicrograph (fig. 13) of a corn root tip which has been exposed to a 1 to 200,000 solution of copper, then colored with  $K_4FeCy_6$ , and sectioned for observation. The dark, abruptly angular line of penetration shown in the section plainly indicates that individual cells may be penetrated by copper while adjacent cells growing under precisely similar physical conditions are not penetrated. If this be not due in some unseen way to morphological peculiarities of root structure, it must be due to individuality in the cells themselves, some of which must be more resistant to penetration by dilute copper solutions than others.

*Summing up the physiological observations relating to effects of copper upon plants*, we find (1) that individual cells vary (probably) in degree of resistance to penetration by copper salts; (2) that young roots are less resistant than old roots; (3) that roots of certain species of plants (e.g. corn) are less resistant than roots of other species; and (4) that toxic effects may be to some extent related to the structure and distribution of root systems.

#### DIAGNOSIS OF COPPER INJURY

In the presence of toxic amounts of copper in the soil, the root systems of culture plants become harsh and crinkly with almost entire loss of root hairs. Consistent with the checking of growing points, root systems are also greatly restricted in extent,

and in feeding capacity. Individual roots are coarse, covered with thick epidermis, and are abruptly angular, apparently as a result of chemotropic contortions. Root tips are shortened and thickened and in some instances are strongly proliferated. The anatomical structures associated with these changes in form are very striking. In corn the cells of the primary cortex, in normal roots, are elongated parallel with the axis of the root, and in longitudinal tangential section measured about 74 by 30 microns. Injured cells of corn grown in soil containing 0.1 per cent copper gave longitudinal tangential sections approximately 34 by 30 microns, as shown on accompanying drawings. (See, also, plates 6, 7, and 8.

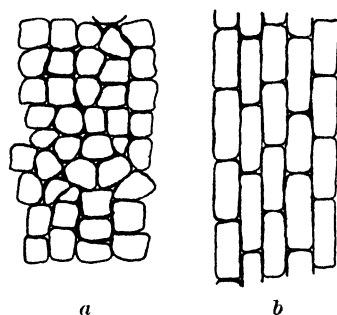


Fig. 14.—*a*. Tangential longitudinal section of corn root grown in soil containing .1 per cent of copper as copper sulphate, showing cells of cortex of injured rootlet. *b*. Tangential longitudinal section of normal corn root cells of cortex. ( $\times \pm 300$  diam.) (Sections by G. F. Freeman.)

These structural modifications, taken in connection with other symptoms and conditions and in the absence of other causes, such as an excess of alkali salts,<sup>9</sup> confirm a diagnosis for copper injury in a soil of doubtful toxicity. For instance, March 4, 1916, two sets of samples of barley were collected in the district studied, and the material examined for evidence of copper injury, as follows:

*Lot 1.*—Young barley plants from the upper end of a field midway between Safford and Solomonville, under Montezuma Canal. The soil next the ditch shows old tailings, and there are irregular areas of yellow barley immediately under the canal.

<sup>9</sup> See Livingston, *Botanical Gazette*, vol. 30, no. 5, p. 229, 1900.

Sample No.	a. Yellow barley plants	
6343	Roots, crinkly and angular, much branched near surface.	
	Dry weight, 3.2429 gms; Cu, .00085 gm; p.p.m. ....	262
6344	Soil shaken from yellow barley roots	
	Copper .....	.07979%
	Total soluble solids (alkali) .....	.46400
	Cl as NaCl .....	.004
	Sodium carbonate .....	.008
	Nitrogen .....	.137
	b. Green barley plants from near (a).	
6345	Roots, smooth and straight, not much branched near surface. Dry weight, 1.6025 gm; Cu, .0002 gm; p.p.m. ....	125
6346	Soil shaken from green barley roots	
	Copper .....	.05844%
	Total soluble solids (alkali) .....	.45600
	Cl as NaCl .....	.004
	Sodium carbonate .....	none
	Nitrogen .....	.181

*Lot 2.*—Young barley plants from the upper end of a field in West Layton under Montezuma Canal. Soil near ditch known to contain tailings and showing spots of yellow barley at head of field.

Sample No.	a. Yellow barley plants	
6347	Roots, crinkly and angular, much branched.	
	Dry weight, 3.2977 gms; Cu, .0014 gm; p.p.m. ....	425
6348	Soil shaken from roots of yellow barley plants	
	Copper .....	.1113%
	Total soluble solids (alkali) ..	.50
	Cl as NaCl .....	.008
	Sodium carbonate ..	.008
	Nitrogen .....	.165
	b. Green barley plants from near (a)	
6349	Roots, smooth, straight, not much branched.	
	Dry weight, 2.2473 gms; Cu, .0003 gm; p.p.m. ....	133
6350	Soil shaken from roots of green barley plants	
	Copper .....	.02678%
	Total soluble solids (alkali) .....	.40
	Cl as NaCl .....	.008
	Sodium carbonate .....	.004
	Nitrogen .....	.127

Considering the above observations, we notice that the soils from which samples were taken do not contain injurious amounts of soluble salts. Their nitrogen content, also, is normal. The areas of yellow barley from which samples come are therefore not to be attributed to alkali salts, or to abnormal nitrogen content. Observation in the field, also, failed to indicate that conditions of irrigation, temperature, or light were unfavorable, these conditions being the same for both green and yellow samples.

Excluding these considerations, therefore, we now find that there is uniformly more copper in the roots of yellow barley plants than in those of the green ones, also in the soils in which they occur. The roots of yellow plants, moreover, show the crinkly condition caused (though not exclusively) by copper when present in toxic amounts in the soil. The following statement summarizes these observations.

Lot 1			
	Cu in soil per cent	Cu p.p.m. in roots	Condition of roots
Yellow barley	.0798	262.00	Crinkly and branched
Green barley	.0584	125.00	Straight, not branched
Lot 2			
Yellow barley	.1113	425.00	Crinkly and branched
Green barley	.0268	133.00	Straight, not branched

The evidence therefore indicates quite conclusively that the two yellow samples owed their color to toxic effects of copper upon the roots of the young plants. Later in the season, however, no difference in mature plants, showing variations in color when young, may be observed. This must be due to the fact that as root systems penetrate more deeply into the soil they escape the surface zone of tailings, with consequent recovery from the effects of copper.

## Part II.—GENERAL DISCUSSION

### PRELIMINARY STATEMENT

The copper compounds, in solid form and in solution, that result from mining operations in the Clifton-Morenci district, have found their way down the San Francisco and Gila rivers to the underlying irrigated agricultural soils of Graham County in sufficient amounts to raise the question of their toxicity to crops. The largest amounts of copper in these soils are found at the heads of irrigated lands, especially where alfalfa is or has been, at which points old accumulations of tailings, laid down for the most part prior to 1908, are still to be found.

### ACCUMULATIONS OF COPPER

The amounts of copper accumulating in the Gila River valley soils in this way are small, the observed range being from 0.006 per cent to 0.111 per cent in surface soils and the average for eighteen soils analyzed being 0.046 per cent of copper. Irrigated soils elsewhere have been observed to contain larger quantities of copper than those above noted, for instance 1.002 per cent on the Deer Lodge River below Anaconda, Montana, with an average of 0.09 per cent for eleven other samples taken in the same locality.<sup>10</sup>

These amounts of copper in a soil may or may not be toxic according to the combination in which the copper exists, the physical character of the soil and its chemical composition, climatic and moisture conditions, the crop grown, and other considerations which may now be discussed in order.

The small amounts of soluble copper constantly coming down stream from the mines which cannot, like solid tailings, be entirely excluded from irrigating water supplies, are of importance because of their tendency to accumulate by reason of the fixing power for copper of silicates, carbonates and organic matter in the soil. The completeness of this fixing power of soil for copper is shown by several experiments in which solutions of copper

<sup>10</sup> U. S. D. A. Bur. Chem., Bull. 113, p. 34, 1907.



salts were percolated through one to twelve inches of soils, with little or no copper appearing in the filtrates. Under field conditions, therefore, this action tends to concentrate dissolved copper in irrigating water in the surface few inches of the soil.

A series of samples of Montezuma Canal water taken at Solomonville affords quantitative suggestions in this connection:

TABLE XXIX  
COPPER CONTENT OF GILA RIVER WATERS

Sample No and date	Description	Amounts of Cu added in irrigation, estimated in p.p.m. of water			Approx flow of Gila River in sec ft	Approx amts of copper carried down stream, 1 day lb.
		In tailings	In solution	Total		
3309 May 26, '04	River very low	18.3	.80	19.1	30	3094
3486 June 11, '05	Small flood	.....	.25		170	230
						(Soluble only)
3622 June 25, '06	River low	1.6	.11	1.71		
3737 Feb. 22, '07	Medium flood	trace	2.88	±3.0	600	9720
4011 Jan. 3, '08		2.1	.08	2.18		
	Tailings shut out of river May 1, 1908					
4029 Apr. 12, '09		1.4	.08	1.48		
6342 Mar. 4, '16*		.04	.03	.07		

\* Following four-months shut-down of operations in Clifton-Morenci district.

These figures, while somewhat meagre, seem to indicate a lessening waste of copper downstream following the restraint of tailings from the water-supply in May, 1908. This is especially true of copper in solution, due probably to the decreased amounts of solid copper compounds in suspension from which copper in solution is derived.

Assuming at the present time an average of 1 part of copper in 1,000,000 of Gila River water, four acre-feet of such water, required for one year's irrigation, would contain 10.9 pounds of copper, from which should be deducted small losses due to vegetation, drainage waters, and percolation to depths below the surface soil.

Six tons of alfalfa with a copper content of 5 p.p.m. contain 0.06 lb. copper; while one acre-foot of seepage water (about the annual seepage loss) containing 0.25 p.p.m. copper would carry 0.68 lb. copper. Estimating the total loss roughly at one pound

of copper per acre a year, the net addition of copper to the soil would be approximately ten pounds, or about 0.00025 per cent. It would therefore require about forty years to accumulate 0.01 per cent of copper in the surface foot of soil. Inasmuch as, under field conditions, this is not an injurious amount, there is little likelihood, considering the district in a general way, that the small residues of copper now coming down stream will accumulate to an injurious extent within a reasonable period of time. Incidentally, it is of interest to note the large total losses of copper (3094 lb. and 9720 lb. per day observed) formerly resulting from mining operations in the district.

#### POSSIBLE EFFECTS UPON HEALTH

With reference to the question of poisonous effects upon man and animals of dissolved copper in irrigating and well-waters, such effects, in general, are much less upon animals than upon plant life. Moore and Kellerman state, for instance, that 0.02 gms. of copper may be absorbed daily by a man with safety.<sup>11</sup> This amount of copper would be contained in five gallons of water containing one part per million of copper, the largest amount of copper observed in a well-water in the district studied being 0.53 p.p.m. It is of interest in this connection to note a belief of the copper miners of the Rio Tinto in southern Spain, where the wells are impregnated with copper, that one part of copper per million of drinking water is permissible, but that two parts per million result in "copper colic."<sup>11a</sup> In view of experiments upon human subjects, however, it is more than likely that deleterious effects observed are due to associated compounds in the water. It is of importance to note that a strength of as little as one part per million of copper in pure water will destroy algae, which are common in clear water supplies freely exposed to light and air. This fact may be made of use in cleaning ditches and reservoirs of aquatic growth, where the expense is not too great.

The germicidal effects of small amounts of copper in waters of the district studied also have a bearing upon human health.

<sup>11</sup> U. S. D. A. Bur. Pl. Ind., Bull. 64, p. 23.

<sup>11a</sup> Conversation of J. W. Bennie, Clifton, Arizona.

Bacilli of various species reacting upon human health are very sensitive to the action of soluble copper salts. For instance, in distilled water "one part copper in 16,000,000 parts water killed typhoid bacilli in two hours. In copper solutions made up with tap and sea water, the action was still marked, but less vigorous than in distilled water."<sup>12</sup> Moore and Kellerman state that one part of copper sulphate to 100,000 parts of water destroys typhoid and cholera germs in three to four hours.<sup>13</sup> In milk supplies as little as one part of copper salts in 2,000,000 of water acts as an antiseptic against putrescent bacteria.<sup>14</sup> It seems, therefore, that there is a possibility that the amounts of copper observed in ditch and well-waters in the district may have an antiseptic effect upon malignant germs, more particularly typhoid fever, likely to occur in the district.<sup>15</sup>

#### AMOUNTS AND SIGNIFICANCE OF COPPER IN AERIAL VEGETATION

The amounts of copper found in aerial parts of vegetation within the district are small, ranging from a trace to 7.6 parts copper in 1,000,000 of dry matter and averaging 3.41 parts. Miscellaneous cultures in water, potted soils, and plots gave larger amounts of copper which, however, were associated in most cases with toxic effects. Table 30 (p. 462) contains a summary of these data.

Even allowing for errors of method and of analysis, the European figures (3) seem excessively high, although the woody character of most of the samples was for the most part very different from that of the tender crop plants of the Arizona series.

Little can be said as to the toxic effects of the copper observed in aerial plant parts in the Arizona samples. The yellow striping of copper-poisoned corn is probably a general symptom of malnutrition to be attributed to the effect of copper upon root systems rather than upon leaves and stems. In rare instances, however, beans and squash in water culture showed dark green

<sup>12</sup> *Biochem. Jour.*, Aug., 1908, pp. 319-323.

<sup>13</sup> *U. S. D. A. Bur. Pl. Ind.*, Bull. 64, p. 43.

<sup>14</sup> *Jour. Ind. and Eng. Chem.*, Sept., 1909, p. 676.

<sup>15</sup> See Bibliography, p. 487, references 3, 20, 21, 22.

TABLE XXX

## SUMMARY OF COPPER CONTENT OF AERIAL VEGETATION

	No of samples	Min.	Max.	Ave.
		Parts per million copper		
1. Field vegetation from upper Gila....	10	trace	7.60	3.41
Field vegetation from other sources in Arizona .....	9	none	6.30	1.52
2. Corn plants grown in pots .01-.05 per cent Cu .....	3	6.5	21.00	13.30
Tops of corn, beans and squash grown in Cu water culture .....	6	11.7	32.00	22.90
Tops of corn, beans, etc., irrigated with copper solutions .....				14.00
Beans in soils containing Cu .....	9	13.0	44.00	26.00
Squash ditto .....	5	14.0	61.00	39.00
Corn ditto .....	20	4.4	239.00	42.00
3. Field samples collected by Leh- mann <sup>16</sup> .....	43	0	560.00	86.00
Field samples collected by Ved- rödi <sup>17</sup>				
1894 .....	26	40.0	1350.00	257.00
1895 .....	26	10.0	680.00	151.00

patches that may possibly have been due to presence of copper, inasmuch as appearances of this character are sometimes noted as an effect of the application of Bordeaux mixture. Bain states, for instance, that extremely minute amounts of copper stimulate formation of chlorophyll in a cell, and therefore increase the formation of starch.<sup>18</sup> Ewart, also, shows that solutions of copper as dilute as 1 to 30,000,000 prevent the action of diastase upon starch.<sup>19</sup> It is possible, therefore, that the juices of plant tissues containing traces to 239 parts (observed) of copper in 1,000,000 of dry matter may carry sufficient of this amount in solution in the cell sap to hinder the action of enzymes upon starch, and thus prevent its normal translocation.

<sup>16</sup> *Der Kupfergehalt von Pflanzen und Thieren in Kupferreichen Gegenden*, Lehmann Archiv für Hygiene, vol. 27, pp. 1-17, 1896.

<sup>17</sup> Quoted in Brenchley, *Inorganic Plant Poisons*, p. 17, 1914.

<sup>18</sup> Bain, Tenn. Agr. Exp. Sta., vol. 15, Bull. 2, p. 93, 1902.

<sup>19</sup> Ewert, Zeitschr. für Pflanzenkrankh., vol. 14:3, p. 135, 1904.

## AMOUNTS AND SIGNIFICANCE OF COPPER IN ROOT SYSTEMS

Of far more and unmistakable importance is the effect of copper on root systems of plants. Under all conditions, whether grown in water culture, in pots, plots, or as field crops, the root systems of plants contain much greater amounts of copper than do the aerial portions, as is shown briefly in the following condensation of results:

TABLE XXXI  
SUMMARY OF COPPER CONTENT OF TOPS AND ROOTS OF PLANTS

	No. of samples	Cu in p.p.m.		Ratio
		Tops	Roots	
Corn, beans, and squash in water cultures, poisoned but living .....	3	22.00	103.00	1 to 4.7
Ditto—killed by copper .....	3	23.00	268.00	1 to 11.6
Corn grown in soil containing .01 per cent of Cu as $\text{Cu}(\text{OH})_2\text{CuCO}_3$ .....	1	6.50	152.00	1 to 23
Corn grown in soil containing .025 per cent Cu as $\text{Cu}(\text{OH})_2\text{CuCO}_3$ .....	1	21.00	728.00	1 to 35
Corn grown in soil containing .05 per cent Cu as $\text{Cu}_2\text{S}$ .....	1	12.50	171.00	1 to 14
Bean series grown in soils containing Cu as pptd. carbonate .0025 to 1.5 per cent Cu in soil .....	9	26.00	1431.00	1 to 55
Corn series grown in soils containing Cu as $\text{Cu}_2\text{S}$ .01 to 1 per cent Cu in soil .....	7	51.00	702.00	1 to 14
Corn series grown in soils containing Cu as chrysocolla, .05 to 1 per cent Cu in soil .....	3	13.00	266.00	1 to 20
Corn series grown in soils containing Cu as pptd. carbonate, .0025 to .05 per cent Cu in soil .....	4	13.00	416.00	1 to 32

Excluding samples grown in water cultures, the roots of which were cleaned with 4 per cent HCl, probably with loss of some copper, the root systems of experimental cultures contained averages of from fourteen to fifty-five times as much copper as the aerial portions of the plants. Furthermore, fine roots of corn were found in one instance to contain about three times as much copper as coarse roots of the same sample, and, finally, the maximum amount of copper, as determined both by analysis and by observation, in water cultures, was found in the root *tips*

of plants affected by copper. Analyses of water cultures of corn, peas, and wheat showing slight toxic effects gave the following ratios of copper in tops, root systems, and root tips:

Water cultures showing slight toxic effects	Cu in p.p.m.		
	Tops	Roots exclusive of tips	Root tips
Corn .....	27.00	91.00	545.00
Peas .....	17.00	1400.00	3428.00
		1680.00	
Wheat .....	130.00	297.00	2811.00

The root tips in this material, by means of caustic potash (the biuret reaction) and potassium ferrocyanide, show the characteristic purple and dark-red reactions due to copper. In the former case not only copper, but copper *in combination with proteids*, is indicated—the purple color being due to the biuret test, which identifies both copper and proteids simultaneously. In roots grown in water culture, and then subjected to the action of dilute copper solutions, the location of copper in a poisoned root system can be seen under a low power with considerable exactness. The purple of the biuret test begins very definitely with the growing point of the root tip and fades out gradually in comet-like fashion usually within one or two millimeters distance of the tip. New growing points in process of pushing their way through the epidermis along the sides of the roots likewise give a strong but very local biuret reaction. This combination of copper (in the form of oxide) and proteids is one used for the precipitation of albuminoid nitrogen in chemical analysis of feeding stuffs.<sup>20</sup> The amount of copper entering into the combination varies with proteids from various sources. As a rule, animal proteids combine with much less copper than vegetable proteids—averaging about 2.4 per cent of copper for egg albumin. Vegetable proteids combine with from 11.60 to 16.97 per cent of copper oxide and average 11.7 per cent copper.<sup>21</sup> Ordinarily, therefore, a vegetable proteid would be saturated with about one-ninth of its weight of copper; but its physiological activities are disarranged and the root killed by much less than the amount required to saturate the proteid.

<sup>20</sup> See Bibliography, p. 488, reference 48.

<sup>21</sup> Mann, *Chemistry of the proteids*, p. 305.

For instance, in samples of wheat and pea roots grown in water culture, it was found by means of nitrogen and copper determinations, using the factor 11.7 per cent copper for saturation of albuminoids, that in wheat roots 4.96 per cent of the copper required for saturation was present and in pea roots 7.99 per cent.

It appears, therefore, first, that copper attacks plant proteids at the most delicate and vulnerable points in the whole plant organization—the growing points of the root systems; and, second, that a small proportion of the copper required for complete reaction is sufficient to kill the protoplasm at these points. Again, it is to be observed that, especially in the seedling stages of growth, the number of growing points is small so that only extremely minute amounts of copper are required to arrest the growth of root tips, the spread of root systems and the nutrition of the plant.

Inasmuch, also, as plants vary greatly in the physical structure and the physiological activity of their root systems, including the number, delicacy and absorptiveness of their growing points, it is not unlikely that the varying sensitiveness to copper salts of different plants, and of the same plant at different ages, may be explained by these observations. Corn, for instance, the most sensitive plant worked with, is characterized in its seedling stages by a small number of vigorously absorptive growing points.

By means of the more delicate dark-red potassium ferrocyanide test, copper may usually be traced through the vessels of the root systems for considerable distances, showing that it is through these channels that small amounts of the metal finally reach the stems and leaves. Here the maximum amounts of copper are found in the outer and upper portions of the plant, where evaporation is most active, and where the greatest residuum of copper therefore occurs. The potassium xanthate (yellow) and hydrogen sulphide (brown) tests also reveal copper in root structures but are not so satisfactory for this purpose as potassium ferrocyanide. (See plate 9.)

The above described reactions, which are so conspicuous in water-culture material killed by copper, are very obscure or imperceptible in roots grown in soils containing copper. The

first material, however, is dead and more nearly saturated with copper; while living roots from soil culture, with proteids combined to but a small per cent of their capacity for copper, do not give satisfactory color tests. These reactions, therefore, do not serve for qualitative determinations of toxic effects in field material.

#### RELATIONS BETWEEN AMOUNTS OF COPPER IN ROOT SYSTEMS AND INJURY TO PLANTS

An effort to establish relations between the amounts of copper in parts per million of dry matter in root systems, and toxic effects as shown in the condition of aerial portions of the plant, was only partially successful; but a sufficient number of observations on samples of sufficient size produced under carefully regulated conditions would probably establish such relations. In the tables shown on the preceding pages there is a fair degree of agreement between the members of each experimental series, the copper found in root systems increasing in most cases with the amount of copper in the soils of each particular series of cultures. In the case of beans and corn grown in cultures containing copper in the form of precipitated carbonate, beans show a somewhat higher resistance to toxic effects and also contain larger amounts of copper in the root systems throughout the series. The conditions under which the samples were grown seem to have, within limits, more effect upon the copper content of root systems than the amounts of copper in the soil, as is indicated in the following tabular statement:

TABLE XXXII

#### TOXIC CONCENTRATIONS OF COPPER IN SOILS AND ROOT SYSTEMS

Culture	Points at which toxic effects begin	Cu in root system at point not showing toxic effects p.p.m.	Cu in root system at point showing toxic effects p.p.m.
Corn, seven samples from field soils .....		42 at .07%	
Corn in field plots containing Cu as sulphate .....	.04%	245 at .025%	296 at .05%
Corn in pot cultures containing Cu as carbonate .....	.023%	748 at .02%	509 at .025%
Beans in pot cultures containing Cu as carbonate .....	.035%	950 at .025%	1358 at .05%



In this statement, for instance, field samples of corn roots grown in soil containing 0.07 per cent of copper contained only 42 p.p.m. of copper in dry matter, while a plot sample grown in soil containing .025 per cent of copper contained 245 p.p.m. of copper in dry matter, and corn grown in pot culture containing 0.02 per cent of copper in soil contained 748 p.p.m. of copper in dry matter.

These differences may be due to the coarser root systems of plot and field-grown samples, this condition being associated with relatively small amounts of copper in dry matter. In view of the great labor involved in preparing root samples for analysis and the very variable results obtained from copper determinations made upon such material, there seems to be little hope of establishing satisfactory ratios of copper to dry matter for the purpose of determining that a sample of field material has been injured by copper. It is probable, however, that for comparative purposes, pot cultures of field soils conducted under uniform and carefully regulated conditions, with standard plants of known behavior, may yield figures of comparative value in determining the character, toxic or otherwise, of a soil containing copper. Corn is an excellent summer-growing plant for the purpose, inasmuch as it shows toxic effects easily, grows rapidly, and affords abundant root materials for analytical determinations. For winter cultures, wheat serves well. Both plants are representative of standard crops for the region under discussion.

#### PATHOLOGICAL EFFECTS

Pathological effects in tops and roots may confirm to a considerable extent, the fact that a plant has been poisoned by copper. The lengthwise yellow striping of corn and wheat leaves due to toxic amounts of copper is not distinctive since the same appearances may result from various other conditions inducing malnutrition, such as those mentioned on a preceding page. Usually, however, careful observation will identify or eliminate these other disturbing factors.

Root systems grown in coppered soils are also conspicuously injured, being stunted in growth, of harsh and crinkly texture

and (in the case of corn) showing characteristic proliferated root tips. The epidermis is thick and rough and the cells in longitudinal tangential section contract from the oblong toward the circular form. Here, again, other factors, such as alkali salts in excess, may lead to similar appearances; and these must be eliminated in a diagnosis of copper injury.

#### SOIL CONDITIONS RELATING TO TOXIC EFFECTS OF COPPER UPON PLANTS

Certain conditions favor, others oppose the toxic action of copper under field conditions, the general tendency being to modify or do away with toxic effects, where the amounts of copper are not excessive.

*Carbon dioxide* in the soil, alone and in conjunction with certain salts ( $\text{NaCl}$ ,  $\text{Na}_2\text{SO}_4$ ) tends to form solutions of basic copper carbonate. Carbonates ( $\text{Na}_2\text{CO}_3$ ,  $\text{CaCO}_3$ ) lessen the solubility of basic copper carbonate in carbon dioxide and, therefore, the toxicity of copper compounds in soils containing these carbonates.<sup>22</sup>

*Coarse, sandy soils* favor toxicity by permitting free movement of solutions and because the withdrawal in them of copper from solution by physical and chemical reactions is minimum.<sup>23</sup>

*The character of the compound of copper* to which roots are exposed is important. In pot cultures of precipitated carbonate of copper, of sulphide in the form of chalcocite pulverized to go through a 100-mesh sieve, and of silicate in the form of chrysocolla pulverized to 100-mesh, toxic effects appeared with corn as follows:

Pot culture of corn; Cu in form of pptd. carbonate—showing toxic effects at .023% Cu in soil

Pot culture of corn; Cu as chalcocite, 100-mesh—showing toxic effects at .08% Cu in soil

Pot culture of corn; Cu as chrysocolla, 100-mesh—showing toxic effects at .08% Cu in soil

The precipitated carbonate is not only more soluble in carbon dioxide than in chalcocite, but also more easily acted upon

<sup>22</sup> See Bibliography, p. 487, reference 12.

<sup>23</sup> See Bibliography, reference 18.

by the acids of plant roots than chalcocite or, probably, chrysocolla. Under field conditions, copper in tailings is originally mostly in the form of sulphides, chiefly chalcocite, which oxidizes only slowly to sulphate in presence of water and air. Chalcocite, 3.2 grams, shaken up with 600 c.c. of water, and air, for twenty-eight days, yielded only 16 mg. of soluble copper. The soluble sulphate in contact with silicates and carbonates of the soil is converted to insoluble forms. The process is gradual and the amount of soluble copper present at any one time is small.

*The tilth of the soil* is significant. A pot culture very thoroughly mixed with 0.1 per cent of copper as carbonate resulted in badly poisoned plants containing about four times as much copper in root systems as in a lumpy mixture of soil containing the same amount of copper. The heavy tailings clay, with which copper is chiefly associated in the district studied, tends to remain in lumps and masses, thus minimizing toxic effects of contained copper compounds.

*In water cultures* toxic effects of copper salts are lessened by salts contained in well-water or in nutrient solutions. This is due, in part, to the presence of other ions, the effect of which is to decrease the ionization of copper salts, with consequent decrease in toxicity. This observation applies to soil-water solutions which contain considerable amounts of alkali salts. It is of interest in this connection to note that certain combinations of salts representing complete mineral nutrients exert maximum antitoxic action to copper salts:<sup>24</sup> and that therefore a fertile soil containing maximum amounts of plant nutrients will tend to minimize toxic effects of copper.

*Antagonistic solutions*, so called, involving copper, may also account for a decrease in toxicity. By reason of a property of the semipermeable membranes of root systems, ions may be either more readily or less readily allowed to penetrate. When penetration is decreased through the addition of ions of other soluble salts this salt is said to be antagonistic in character. Copper is thus antagonized by sodium and potassium salts, of which the soluble salt content of the soil is chiefly composed.<sup>25</sup>

<sup>24</sup> A. Le Renard, *Essai sur la valeur antitoxique de l'aliment complet et incomplet*. Abstracted in *Science* n. s. vol. 28, no. 712, p. 236, 1908.

<sup>25</sup> See Bibliography, p. 488, references 35-44, 52.

*Physical attractions*, or adsorptive effects, also account for a very considerable lessening of the amount of dissolved copper salts, in contact with soil particles. Jensen, for instance, finds that a dilute copper solution is ten times as toxic in the free condition as when it is mixed with an artificial quartz soil, that is to say, the quartz reduces the toxic effects about nine-tenths. Inasmuch as the reduction in toxicity is a function of the solid surface to which the soluble salts are exposed, the finer the state of division of a soil the more will be the adsorption and the less will be the toxic effects of a stated copper solution.<sup>26</sup>

*The age of plant roots* markedly affects their susceptibility to copper salts. Young and tender roots, containing large amounts of protoplasm, are much more quickly and easily poisoned than old and comparatively fibrous structures containing a small proportion of protoplasmic materials. This may be due to differences in the thickness of cell walls protecting the cell contents from outside substances; it may be due to a different degree of permeability of the protoplasm of older roots to copper salts; or it may be due to lessened reactivity due to changed chemical character. In any case, this observation indicates a distinctly greater resistance to copper in soils, of older, more fibrous, and possibly intrinsically more resistant root systems. Different species of plants also show varying degrees of resistance to copper salts. In pot cultures, peas are distinctly more resistant to precipitated carbonate of copper than corn. Different plants of the same species also show a certain amount of individuality with reference to absorption of copper.

### STIMULATION

Not only do the various influences described above lessen the toxic effects of copper upon plants, but it is possible, also, that the amounts of copper may be decreased in the field to the point at which stimulating effects occur. As shown in the discussion of water cultures on preceding pages, extreme dilutions of copper salts in distilled water, for instance, 1 part to 100,-

<sup>26</sup> G. H. Jensen, *Botanical Gazette*, vol. 43, p. 11, Jan., 1907.

000,000, caused increased growth of root tips growing in these solutions. This observation accords with those of some other experimenters, not only with copper solutions but with solutions of various other metals, and bears a certain analogy to stimulating effects upon animals observed with very small amounts of poisons, such as arsenic and strychnine. Stimulation was also observed in the case of certain pot cultures watered with dilute copper solution in such a way that these solutions were filtered through a thin layer of soil before they reached the plant roots. Under these conditions a portion of the root systems must come in contact with extremely dilute copper solutions residual from the reactions of copper salts with the soil. As in the case of water cultures, these extremely dilute solutions must have exerted the stimulating effects which were apparent in several cultures made in this manner.

In the case of pot cultures also, in which stated amounts of copper were uniformly mixed throughout the soil, apparent stimulation of growth was occasionally observed; for instance, with 0.01 per cent of copper in the form of precipitated carbonate in a culture of corn.

A satisfactory explanation of stimulation effects is not available. It is to be supposed that stimulation in a soil culture in which copper sulphate is used may be explained by the action of the  $\text{SO}_4$  ion upon the soil in releasing plant food for the use of the plant. However, such stimulation is seen in water cultures where this does not occur. Lipman<sup>27</sup> has observed that under certain conditions the nitrifying flora of soils is stimulated by salts of copper, zinc, iron and lead. Such stimulation, through increased elaboration of nitrates, may account for the behavior of cultures showing increased growth. Stimulation effects, therefore, which undoubtedly occur both in water and in soil cultures, are perhaps due to more than one different cause—to chemical and bacterial agencies in soils, and to a pathological disturbance in water cultures.<sup>28</sup>

Taking into account the very minute amounts of copper salts with which stimulated growth is associated, and the very gradual

<sup>27</sup> Lipman, C. B., and Burgess, P. S., Univ. Calif. Publ. Agr. Sci., vol. 1, no. 6, pp. 127-139, 1914.

<sup>28</sup> See Bibliography, pp. 487-488, references 2, 4, 27, 33, 53, 45.

addition of copper to new ground that may occur through irrigating waters, it is not impossible that in favorable situations an actual increase in vegetable growth in the field due to copper may take place; but it is not possible in the field to prove this supposition because of many other factors, the effects of which prevent trustworthy observation.

#### FIELD OBSERVATIONS

In view of the many factors influencing results in the field, some leading towards toxic copper effects, some opposing toxic effects, and still others pointing to the possibility of stimulated growth, it is of interest, finally, to refer to field conditions as they have existed in irrigated lands under the Clifton-Morenci mines for the twelve years during which the district has been under observation. At the beginning of this period, in 1904, considerable accumulations of copper-bearing tailings were evident, more particularly at the heads of alfalfa fields, where they sometimes attained a thickness of as much as ten inches or more. These blankets usually thinned out and disappeared between 100 and 200 feet from the head ditches, leaving crops in lower portions unaffected. Deposits of river sediments were observed in other irrigated districts not affected by mining detritus. The growth of alfalfa was more depreciated by the denser and thicker tailings blankets; and yellow foliage of young grain and young corn was considerably in evidence in tailings, but not as an effect of ordinary sediments. In 1908, the tailings were impounded, and some of the best farmers began the practice of cultivating alfalfa to break up the old accumulations, incorporate them with the soil, and secure better penetration of water and air to the roots of the crop. Following this procedure the stunted growth at the heads of alfalfa lands has considerably but not yet entirely recovered. Patches of yellow young barley, wheat, and oats are still to be observed on old tailings deposits; but as the plants become older they become normal in appearance, and yield apparently normal crops. These observations, which may be repeated many times in the course of a day's reconnaissance in the district, from May to September for alfalfa, and February

to May for grain, may be explained by the following considerations: The wedge-shaped deposit of tailings indicated in the diagram (fig. 15) at first so obstructed access of water and air to alfalfa root-systems that only stunted development was possible either of roots or tops. With an annual cultivation of this blanket and the incorporation of river sediments and better penetration of irrigating waters, deleterious effects tend to disappear and the crop again approaches normal.

Similar land when plowed for grain contains most of the copper associated with old tailings at the surface of the soil. Young grain, therefore, with shallow and susceptible root sys-

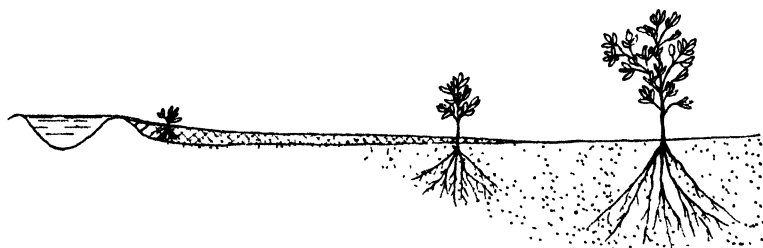


Fig. 15.—Diagram showing behavior of root systems under influence of tailings blanket.

tems, at first, if ever, shows effects of copper in the soil, recovering as root systems penetrate to greater depths where they encounter uncontaminated soil.

#### EFFECTS OF RIVER SEDIMENTS

With reference to the further trend of copper effects upon vegetation in the district, assuming the permanent exclusion of solid tailings but a constant addition of about one part of copper to one million of irrigating water used, it is of interest to take into account the diluting effect of river sediments upon copper compounds in the district.

In four acre-feet of Gila River water, these sediments will amount to about eighty tons per acre a year,<sup>20</sup> of which amount the ten pounds of copper contributed in irrigating waters is only 0.006 per cent.

<sup>20</sup> Forbes, R. H., *Ariz. Agr. Exp. Sta. Bull.* 53, p. 61.

Irrigating sediments alone, therefore, considered in their general relation to amounts of copper which cannot be prevented from reaching irrigated fields, are sufficient in quantity to reduce ultimately the amounts of copper observed below 0.01 per cent in the soils of this district. Since 0.01 per cent is a safe minimum, river sediments, alone, incorporated with the soil are probably sufficient to ameliorate gradually existing accumulations of copper salts and to take care of further contributions in soluble form which cannot at present be avoided.

#### EFFECT OF CULTIVATION UPON ALFALFA

Finally, it is of interest to observe the improvement in a field of alfalfa, in the district studied, between the years 1905 and 1916.

June 23, 1905, the writer carefully measured, cut and weighed a representative plot of alfalfa in William Gillespie's field near Solomonville, Arizona. This field was suffering from an accumulation of tailings, the depreciation in yield at the upper ends of alfalfa lands being conspicuously evident. Following the exclusion of tailings from the irrigating supply in 1908, and with a cultivation each winter with a disk or a spring-tooth harrow, the condition of the field gradually improved until, June 13, 1916, the writer returned and again measured, cut, and weighed the identical plot of alfalfa that had shown bad effects eleven years before. Following are the data, with diagrams, relating to these two cuttings of alfalfa, which are representative for the district within which tailings were deposited.

##### 1. *Alfalfa seriously affected by tailings, June 23, 1905.*

Three lands in William Gillespie's field east of house, near Solomonville, under Montezuma Ditch, out of Gila River. A good stand of alfalfa five years old. Heavy adobe soil; field never disked.

The three lands observed were, over all, 95 feet wide, and divided into plots 100 feet long from top to bottom of field. Ten feet next the ditch was discarded because of banks and bare spots, and the extreme lower portion of the field because of roadways. A portion of plots 6 and 7 was discarded on account of Johnson grass.



Observations were made June 23, 1905, on the second cutting, just beginning to bloom, the field having been irrigated twice since the last cutting. After stirring and raking, the yield of dry hay was weighed June 24. Weather very hot and dry. Following are the data relating to this series:

Plot	Dimensions in feet	Height of alfalfa, inches	Yield of plot, pounds	Tons per acre	Depth of tailings on plot, inches	Condition of surface soil at time of cutting
1	95x100	19	240	.69*	1½-3½	Dust-dry and somewhat cracked
2	95x100	20	340	.87*	1-2	Dry and badly cracked
3	95x100	23-25	570	1.31	¾-1½	Dry, cracked at upper end
4	95x100	24	595	1.36	½-1	Moist, not cracked
5	95x100	23	550	1.26	¾-1	Moist, not cracked
6	60x100	28	400	1.41*	½-1	Moist, not cracked
7	60x100	27	430	1.48*	¾- 1	Moist, not cracked

\* Corrected for thin stand and trash.

## 2. *Alfalfa slightly affected by tailings, June 13, 1916.*

The same three lands, continuously in alfalfa since 1905. A perfect stand, thin spots reseeded by means of a seed crop in 1915. The field had been spring-tooth harrowed each winter for about ten years, especially at heads of lands, to break up the tailings blanket and secure better penetration of irrigating water.

As in 1905, ten feet next the ditch was discarded, also the extreme lower portion of the field. Johnson grass had nearly entirely disappeared.

Observations were made June 13, 1916, on the second cutting, just beginning to bloom, the field having been irrigated twice since the last cutting. After raking and piling, the dry hay was hauled and weighed June 17. The weather was moderately hot and dry; and conditions generally the same as those under which the crop was cut in 1905. Following are the data for the second series of observations:

Plot	Dimensions in feet	Height of alfalfa, inches	Yield of plot, pounds	Tons per acre	Appearance of tailings	Condition of soil at time of cutting
1	95x100	36-21	875	2.00	Distinct	Surface drier soil
2	95x100	22-34	857	1.96	Distinct	Surface dusty, drier soil
3	95x100	34-36	972	2.22	Slight	Moist throughout
4	95x100	30-36	910	2.08	None	Moist throughout
5	95x100	31-33	900	2.05	None	Moist throughout
6	95x100	28-36	860	1.96	None	Moist throughout
7	95x100	34-37	870	1.98	None	Moist throughout
8	95x100	33-36	910	2.08	None	Moist throughout

Comparing these two statements, and illustrating them by means of the following diagram (fig. 16), it is evident that the depreciation in yield observed in the upper plots in 1905 has disappeared in 1916, the yields on the last date being practically uniform from top to bottom of the field. Effects of tailings are still plainly visible in plots 1 and 2 in spots and patches of short alfalfa, compensated for, however, by areas of stimulated growth apparently due to seepage from the adjacent ditch. The yield of the field as a whole is also much improved due to cultivation and reseeded of the field.

In brief it may now be stated that, following the exclusion of tailings from the irrigating waters of this locality, it has been found possible, in this carefully observed case, to overcome the deleterious effects of tailings deposits upon alfalfa, slowly but almost entirely, in about ten years.

Thus, co-operation between miners, in restraining tailings from irrigating streams, and those farmers who cultivate their alfalfa intelligently, effectually disposes of the most serious problem that has arisen in connection with copper-mining detritus.

The chemical composition of tailings, in fact, would indicate that, as in the case of humid region subsoils, when they are enriched by the addition of organic matter and nitrogen, and filled with bacterial life, they may make very good soil. Following is a statement of the composition of four representative samples of ores and tailings, with reference to potash and phosphoric acid:

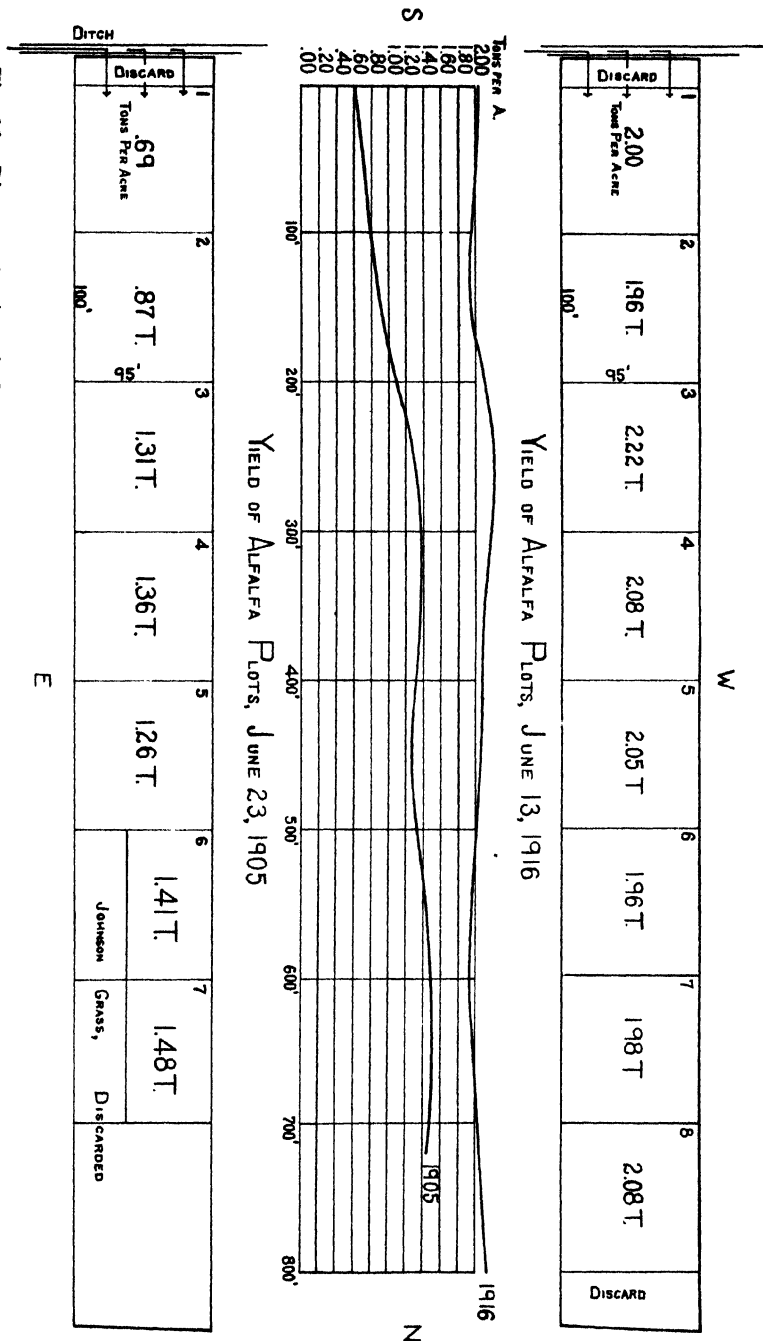


Fig. 16.—Diagram showing yields of alfalfa from head to foot of a land damaged by tailings in the field of Wm. Gillispie, east of house, June 23, 1905; and the same land June 13, 1916, tailings having been kept out of the water since 1908, with the field disk harrowed annually.

Sample No.		Potash $K_2O$	Phosphoric acid $P_2O_5$	Nitrogen N
3491	Sulphide ore	.64%	.11%	Doubtful
3492	Oxidized ore	.44	.11	
3438	Sulphide tailings	.79	.29	traces
3439	Oxidized tailings	.67	.12	

These ores and the tailings derived from them are rich in potash, and contain unexpectedly large amounts of phosphoric acid; but nitrogen is almost nil.

### SUMMARY

1. Copper is shown, as a direct effect of the Clifton-Morenci mining operations in Arizona, to be distributed throughout water-supplies, soils, the vegetable and the animal life of an underlying irrigated district.

2. Smaller amounts of copper are found elsewhere in the State where the drainage basin includes mining operations or ore-bearing areas.

3. Individual plants grown in water cultures or in soil containing copper show a comparatively small, and probably not injurious, accumulation of copper in the aerial portions of the plants; but the root systems, carefully cleansed of externally adhering copper, contain relatively great amounts.

4. Copper in root systems, as shown by the biuret test, is largely in combination with plant proteids, especially at the growing points of root systems and near vicinity. The place and nature of the reaction accounts for the extreme toxicity of copper salts to plants. The varying sensitiveness of plants to copper salts may possibly be explained in part by the number and disposition of exposed growing points.

5. Conditions favoring toxicity of copper compounds are the presence of carbon dioxide and certain soluble salts which assist in forming copper solutions that come into contact with plant roots; coarse, sandy soils favoring free access of copper solutions to plant roots and minimizing the withdrawal of copper from solution by adsorption; and the presence of copper in the form of the more soluble precipitated carbonate.

6. Conditions opposing toxicity of copper compounds are the presence of copper in the form of chrysocolla and chalcocite; adsorption through contact with finely divided soil particles; reactions with carbonates, silicates, and organic matter tending to precipitate copper from its solutions; the presence of certain soluble salts in the soil that overcome toxic action; and increased resistance of old plant roots.

7. The stimulation by copper of vegetative growth in pot and water cultures has been observed. Stimulated growth of crops under field conditions is a possibility.

8. Pot cultures may be used for comparative determinations of toxic effects upon plants of copper in soils, if conducted under rigidly uniform conditions. The copper content and the physiological response to copper of such material will be much greater than for similar cultures grown under plot or field conditions.

9. Copper injury in field soils containing doubtfully toxic amounts of copper may be diagnosed by a combination of symptoms. Facts which indicate such injury in a soil containing 0.1 per cent of copper (more or less) are: yellow tops (for winter grains) in absence of other conditions that cause yellow tops; crinkly root systems (in absence of excessive amounts of alkali salts); and a high copper content in dry matter of root systems. Combined evidence of this character, which may be observed in the district studied, indicates toxic copper effects.

10. Field observations before and following the exclusion of tailings from the irrigating water-supply indicate that conditions in the district studied are gradually improving, due to the cultivation of alfalfa and to the incorporation of river sediments with accumulations of tailings. Noticeable toxic effects in the field exist only where the roots of young, growing crops are exposed to surface soils containing maximum amounts of copper. The general tendency in the district is probably toward decreasing rather than increasing percentages of copper in irrigated soils.

11. Methods of analysis have been developed for the purpose of determining reliably small amounts of copper in vegetative material, particularly in root systems of plants grown in soils containing copper.

## Part III.—APPENDIX

### METHODS OF ANALYSIS

WITH THE COLLABORATION OF E. E. FREE AND DR. W. H. ROSS

Freedom of samples, especially vegetation, from contamination with adhering copper; and accurate methods for determining minute amounts of copper in sediments, soils, waters and vegetation, are vital to the integrity of the work recorded in this publication.

Unusual care was taken to perfect methods for preparation of samples, especially roots grown in media containing copper; and refined manipulation in the determination of copper reduced the limit of error to approximately .00001 gram, or .01 milligram.

### REAGENTS AND APPARATUS

*Distilled water* of three derivations was used: (1) University of Arizona well water very slowly distilled through a block-tin worm; (2) the same, redistilled from glass; and (3) University of Arizona well water distilled from glass.\*

*Nitric and sulphuric acids* from Baker & Adamson were used.

*Ammonia and H<sub>2</sub>S* employed were passed through two wash bottles.

Blank determinations from time to time with reagents employed gave no trace of copper, thus insuring results obtained by means of them.

Copper was determined by electrolysis, in minute amounts according to the manipulation of E. E. Free.<sup>1</sup>

The balance used was a No. 2112 Eimer and Amend short-beam assay balance, "distinctly sensitive to 1/200 milligram."

### MANIPULATION

*Ores and tailings.*—1–2 gms. were digested with a mixture of 8 c.c. HNO<sub>3</sub> and 5 c.c. HCl on a hot plate, then 4 c.c. H<sub>2</sub>SO<sub>4</sub> added and evaporated to H<sub>2</sub>SO<sub>4</sub> fumes (method used in Old Dominion laboratory at Globe, and Copper Queen at Bisbee). Took up with water, filtered, neutralized with ammonia, then added 2 c.c. H<sub>2</sub>SO<sub>4</sub> and a few drops of HNO<sub>3</sub> and electrolyzed.

*Soils.*—Soils were examined by two methods:

(a) 100 gms. soil was treated with a mixture of 80 c.c. HNO<sub>3</sub> and 20 c.c. H<sub>2</sub>SO<sub>4</sub> and digested in a porcelain dish on a hot plate to sulphuric fumes; digested with 200 c.c. water, filtered, washed up to about 500 c.c., evaporated to 200 c.c. precipitated iron with ammonia, filtered, washed with about 500 c.c. water, alkaline filtrate reduced by evaporation, acidified faintly with HCl and H<sub>2</sub>S passed for half an hour. The faint black precipitate was

<sup>1</sup> Electrolytic determination of minute quantities of copper, 12th Gen. meeting Am. Electrochem. Soc., October 17–19, 1907.

allowed to settle several hours, then filtered, and the precipitate, including filter, digested with 5–10 c.c.  $\text{HNO}_3$  and water until copper was dissolved, solution filtered, a few drops of  $\text{H}_2\text{SO}_4$  added, evaporated to fumes, and copper determined by electrolysis with addition of 5–25 drops of  $\text{HNO}_3$ .

(b) 200 gms. soil was digested as above with  $\text{HNO}_3$  and  $\text{H}_2\text{SO}_4$ , evaporated to fumes of  $\text{H}_2\text{SO}_4$ , digested with water, filtered and washed up to 500 c.c., made alkaline with ammonia and made up to 1000 c.c. After settling, 500 c.c. or 100 gms. aliquot, was filtered off and copper determined as in (a).

*Waters.*—Waters were evaporated to dryness, the residue digested with sulphuric acid and water, filtered hot, excess of  $\text{H}_2\text{SO}_4$  evaporated, filtered into platinum dish, a few drops of  $\text{HNO}_3$  added, and electrolyzed.

*Vegetation.*—Air-dried samples were burned in a small sheet-iron stove, the iron of which was found to contain *no trace* of copper. Two samples of mistletoe, difficult to burn, were reduced in a *new* muffle in gasoline assay furnace. The charred and partly burned material was moistened with water, and concentrated  $\text{HNO}_3$  added (100 to 200 c.c.) until effervescence ceased, digested until in plastic condition, diluted with hot water and filtered. Evaporated bulky filtrate to dryness, took up with water and  $\text{HNO}_3$ , filtered (getting rid of much organic matter), added about 20 c.c.  $\text{H}_2\text{SO}_4$ , evaporated to  $\text{H}_2\text{SO}_4$  fumes, driving off all but about 5 c.c.  $\text{H}_2\text{SO}_4$ , added water, filtered off insolubles, made up filtrate to about 500 c.c., passed  $\text{H}_2\text{S}$ , and proceeded as usual for copper.

The completeness of the extraction of copper from vegetation by the above method was verified as follows: The extracted, charred residue from 2 lb. 8 oz. of dry corn leaves and blooms in which 1.32 parts Cu per million was found (Sample 3529) was removed from filter paper after washing, moistened with  $\text{H}_2\text{SO}_4$  and additionally burned in a porcelain dish, being finally reduced, after again moistening with  $\text{H}_2\text{SO}_4$ , in a platinum dish in the muffle. The resulting pink ash was then fused with three parts of dry  $\text{Na}_2\text{CO}_3$  (Kahlbaum) and poured on clean porcelain. The fusion was soaked in water with addition of  $\text{H}_2\text{SO}_4$ , evaporated nearly to dryness, filtered from insoluble portion (lime, salts, etc.), again evaporated and filtered, and a third time the same, finally driving off excess of  $\text{H}_2\text{SO}_4$  and electrolyzing as usual. A black precipitate of carbon but *no Cu* was obtained, the same being true of a blank determination on the  $\text{Na}_2\text{CO}_3$  used.

Roots of plants grown in water cultures or in soils must be most thoroughly cleansed of externally adhering copper, since this will introduce excessive errors where the content of copper is small. Three methods of preparing roots for copper determination were employed:

1. Roots grown in water cultures containing copper were dipped for about ten seconds in 4 per cent  $\text{HCl}$ , immediately

washed in copper-free water and dried. Careful observation indicated that adhering copper salts deposited from water solution were completely removed by this treatment. It is probable that the acid penetrates plant tissues somewhat in the time employed and removes some copper. The results are, therefore, probably severely conservative.

2. Roots grown in soil cultures containing copper cannot be safely cleansed with HCl, which does not readily dissolve silicates and sulphides of copper, and which cannot be allowed to remain in contact with plant roots for more than a few seconds.

Carbon dioxide in water was finally selected as a mild, slow but finally effective solvent for the purpose. Samples of roots were first very thoroughly washed in copper-free well-water, then placed in five-liter jars with ground glass covers, a stream of washed CO<sub>2</sub> passed, the jars shaken and treatment with CO<sub>2</sub> repeated until the water was saturated, then allowed to stand with occasional shaking for twenty-four hours. The solution was then siphoned or filtered off and the treatment repeated until, on evaporating the bulky filtrates, no more copper was found. To prevent putrefaction during long-continued washings, a pinch of thymol was added to each washing. From nine to thirty-one washings were found necessary to cleanse plant roots thoroughly, the process being laborious and time-consuming. When the sample yielded no more copper to wash waters it was dried, burned and copper determined according to the method for small amounts in plant ashes.

Following is a record of washings for examples of roots cleaned by this process:

(1) Corn roots grown in a pot culture of soil containing 0.01 per cent of copper as basic carbonate.

	H <sub>2</sub> S test		Quantitative by electrolysis
First wash	distinct		
Fifth wash	distinct		
Ninth wash	doubtful	1 liter of filtrate	no Cu

(2) Corn roots grown in a pot culture of soil containing 0.05 per cent copper as Cu<sub>2</sub>S.

		Quantitative by electrolysis
Tenth wash	2 litres of filtrate	.00006 gm. Cu

(3) Barley roots from field soil containing tailings.

		Quantitative by electrolysis
First wash	2.433 litres of filtrate	.00035 gm. Cu
Second wash	2.531	.00012
Fifth wash	2.22	.00009
Sixth wash	2.41	.00004
Seventh wash	2.00	.00002
Eleventh wash	2.00	.00000



(4) Coarse roots of field corn grown in soil containing tailings.

	H <sub>2</sub> S test	Quantitative by electrolysis
Twenty-fifth wash	distinct	
Twenty-ninth wash		.00005 gm. Cu
Thirty-first wash		.00000

Samples vary as to number of washings required to remove the last trace of copper, but the definiteness with which, finally, copper usually ceases to be extracted by CO<sub>2</sub> water indicates completeness of the operation. This is further emphasized by the comparatively large amounts of copper which are then found in root systems thus cleansed.

3. A third method of preparing roots for copper determination, involving less labor than by washing in CO<sub>2</sub> water, is as follows: Cleanse roots thoroughly in clean water with a camel-hair brush, dry, burn and weigh the ash, then estimate total copper. Determine copper in soil shaken from sample, assume ash as all soil and deduct copper in this amount of soil from total copper found in ash. Results by this method are low, but not seriously in error if sample is thoroughly washed.

Example	Dry matter	Ash	Gms. Cu	Pts. Cu per million
Sample 2a grown in soil containing 0.05% copper	.3561 gm.	10.84%	.000115	322
Ash in sample	.0386			
Copper in ash assumed as soil			.000019	
Net copper assumed			.000096	270

The correction introduced reduces parts per million of copper from 322 to 270, which latter figure is conservative in character.

Of the three methods above described, No. 2 is undoubtedly most exact, but is extremely laborious and time-consuming.

#### THE DETERMINATION OF COPPER IN SMALL AMOUNTS OF PLANT ASHES

The ash is placed in a platinum dish without previous pulverization and moistened with concentrated sulphuric acid in sufficient quantity to bring all parts of the ash in intimate contact with the acid. The material is then thoroughly stirred and heated on a sand bath until fumes of SO<sub>3</sub> begin to come off, then allowed to cool and a sufficient quantity of hydrofluoric acid added to bring the acid in contact with the whole mass, then allowed to stand for at least half an hour and again heated until

$\text{SO}_3$  fumes come off. The material is now washed into a casserole, moistened with sulphuric and nitric acids and digested at a low heat for at least one hour. The heat is then increased until  $\text{SO}_3$  fumes are again driven off. The mass is moistened with three to four times its bulk of distilled water and digested at a gentle heat from one to two hours, filtered hot and then the filtrate and washings evaporated almost to dryness, thus driving off the excess of sulphuric acid. The resulting residue is taken up with hot water and again filtered to separate the solution from precipitated calcium sulphate. This evaporation and filtration may have to be repeated one, two or three times in order to get the solution sufficiently free from calcium sulphate. The final filtrate, which contains the copper, is then diluted to about 150 to 200 c.c. in a tall beaker, a small quantity of hydrochloric acid is added and hydrogen sulphide passed until the solution is thoroughly saturated. During the hydrogen sulphide precipitation there should be no nitric acid or nitrates present in the solution. A large quantity of organic matter is also disadvantageous and may be avoided by evaporating the solution several times to dryness with nitric and sulphuric acids, finishing finally with an evaporation with sulphuric acid alone in order to drive off all traces of nitric acid.

The precipitate from the treatment with hydrogen sulphide is filtered off, washed with water saturated with hydrogen sulphide and digested with a small quantity (2 to 5 c.c.) of nitric acid in a casserole. The digestion should be begun cold and the heat gradually increased. If the digestion is begun at a high temperature the sulphur formed by the decomposition of the copper sulphide will form a film of molten sulphur around the granules of copper sulphide, and this tends to prevent their solution in nitric acid. The precipitate after digestion in nitric acid should be a clear green or else a yellow. If there is any trace of dark color, brown or black, it means that either organic matter has been precipitated with the copper sulphide precipitate, which is extremely unlikely, or else that the above-mentioned sulphur film has formed around some of the particles of copper sulphide preventing their solution in the nitric acid. If the latter be the case, the determination may still be saved by placing the precipitate in a platinum dish and heating over a gentle flame until the sulphur is volatilized. The residue of copper sulphide or of copper oxide may then be digested in nitric acid. The digestions in nitric acid should not be carried to a heat high enough to decompose the copper nitrate formed by the solution of copper sulphide.

After digestion in nitric acid and the evaporation of any large excess of nitric acid, the residue is taken up in hot water, acidified to contain 2-4 per cent nitric acid and filtered into a large platinum dish,  $\frac{1}{4}$  to  $\frac{1}{2}$  c.c. of sulphuric acid is added, and the solution electrolyzed with a voltage of from 2 to  $2\frac{1}{2}$

volts and a current not greater than one ampere. The voltage may be higher than  $2\frac{1}{2}$  volts if necessary but should not be high enough to raise the current beyond the limit given. The electrolysis should be continued at least three hours and preferably nine to twelve hours. The dish is, of course, the cathode. When the electrolysis is complete the electrolyte is washed out of the dish by means of the sucking-bottle and the dish is thoroughly washed with distilled water. In case the deposit of copper on the dish is spongy and loosely adherent it is not safe to wash out the electrolyte. In this case the copper should be redissolved and the electrolysis repeated, using a little more sulphuric acid. If the copper still refuses to come down in adherent form the addition of 2 to 5 c.c. of a one per cent solution of gelatine will often assist the precipitation. In case of a stubborn refusal of the copper to give an adherent deposit it is necessary to dissolve it, evaporate to dryness with sulphuric acid, and reprecipitate with hydrogen sulphide, continuing the process from this point as before.

If the copper refuses to come down at all the trouble is probably an excess of acid in the solution. This may be corrected by the addition of a few drops of ammonia. The concentration of acid in the solution must lie between one and five per cent. At least a small part of this should be sulphuric acid as nitric acid will be destroyed in the course of the electrolysis if it alone is present, and the solution may become alkaline (from  $\text{NH}_4\text{OH}$ ), which will prevent proper precipitation. Chlorides and organic salts, such as acetates and tartrates, should be carefully avoided.

The resulting deposit of copper will probably contain traces of carbon and possibly of platinum. In order to eliminate these and at the same time precipitate copper upon an electrode more suitable for accurate weighing, a second electrolysis is made, using this time the dish as anode and using as cathode a small spiral of platinum wire suspended from a hook of silver (or platinum) wire which in turn is connected to the battery. The electrolysis should also be conducted in nitric and sulphuric acid solution and what is said above as to obtaining satisfactory deposits applies with equal force here. In this case, however, owing to the small surface area of the cathode, it is necessary to work with very much smaller currents than were used in the first electrolysis. The maximum current to be used must be so adjusted by trial as to give bright and adherent deposits. 1-100th ampere and 1.8 volts is a good current for the purpose. It is well to use as the source of current for this electrolysis four Edison-Lalande cells and to have in the circuit a resistance of from 30 to 80 ohms. This gives an electromotive force at the dish of about 1.8 volts. Two determinations may be run in parallel. In this case it is not permissible to use a gelatine solution in order to secure satisfactory deposits, as the copper will be slightly contaminated with gelatine and the obtained weight will be too high.

The electrolysis should be run at least nine hours. When completed, the electrolyte should be washed out as before without breaking the current, the electrode lifted from the solution, disengaged from the supporting hook, and washed and dried by dipping successively in water, alcohol and ether and placing in a desiccator over sulphuric acid. After having remained in the desiccator for an hour the electrode is ready for weighing. Weighings should be made on an assay (button) balance adjusted to maximum sensibility. After weighing, the copper is removed from the electrode by dipping in concentrated nitric acid, and the electrode cleaned and dried by dipping successively in distilled water, alcohol and ether and placing in a desiccator. It is again weighed as before and the difference of the two weights gives the copper obtained.

The electrolyte (from each electrolysis) which has been washed out of the dish by means of the suction flask, is evaporated to dryness taken up with water, acidified with nitric acid and tested for copper by electrolyzing, using the point of platinum wire as cathode. In this way any possible loss of copper by incomplete precipitation in either of the electrolyses is prevented. If any copper is found in this check test it should be dissolved from the platinum wire, added to the solution obtained by dissolving the copper from the small electrode, and the electrolysis repeated in order to get the true weight.

In case a quantity of copper too small to be weighed is obtained its identity as copper may be most easily established by electrolyzing it onto the point of a platinum wire as described above. In these electrolyses with the platinum wire as cathode the current must, of course, be kept low in order to obtain satisfactory deposits. If this precaution is observed the deposit on the platinum wire will be of a brilliant red color and easily distinguishable as copper. If the deposit is brownish or blackish its identity as copper may be established by the green flash when the point of the wire is held in the colorless flame of the Bunsen burner, particularly if the wire has been first dipped in hydrochloric acid. Nitric acid must not be used, as nitric acid itself will give a green flash in the Bunsen burner flame.

The reagents used in the above process should all be tested as to freedom from copper. The water used should be doubly distilled and, at least the second time, from glass. All utensils should be cleaned by boiling in nitric acid. Care must also be taken to conduct the operations in rooms free from dust which might possibly contain copper.

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**PLATE 6**

**Fig. 1.**—Root system of corn plant injured by 0.1 per cent of copper added as copper sulphate to the soil.

**Fig. 2.**—Normal corn root grown in similar soil containing no copper.  
(Photos by G. F. Freeman.)



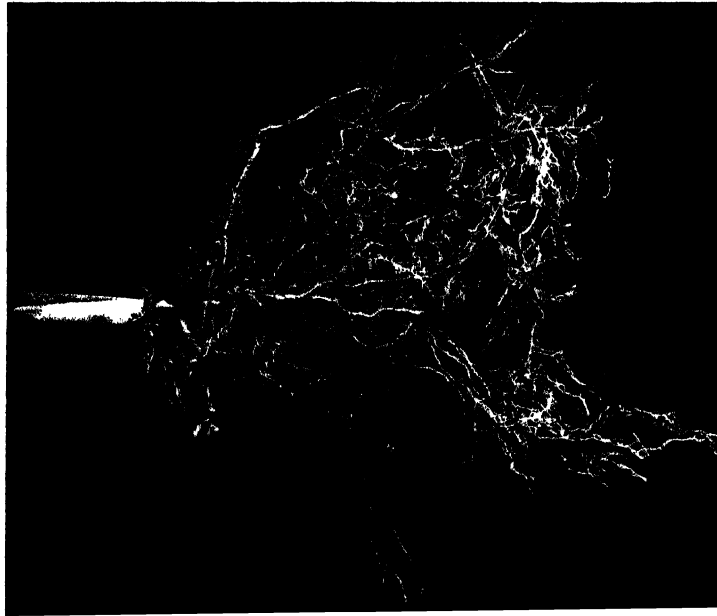


Fig. 1



Fig. 2

### PLATE 7

Fig. 1.—Individual roots of corn injured by 0.1 per cent of copper added as copper sulphate to the soil.

Fig. 2.—Individual root of corn, normal.

(Photos by G. F. Freeman.)

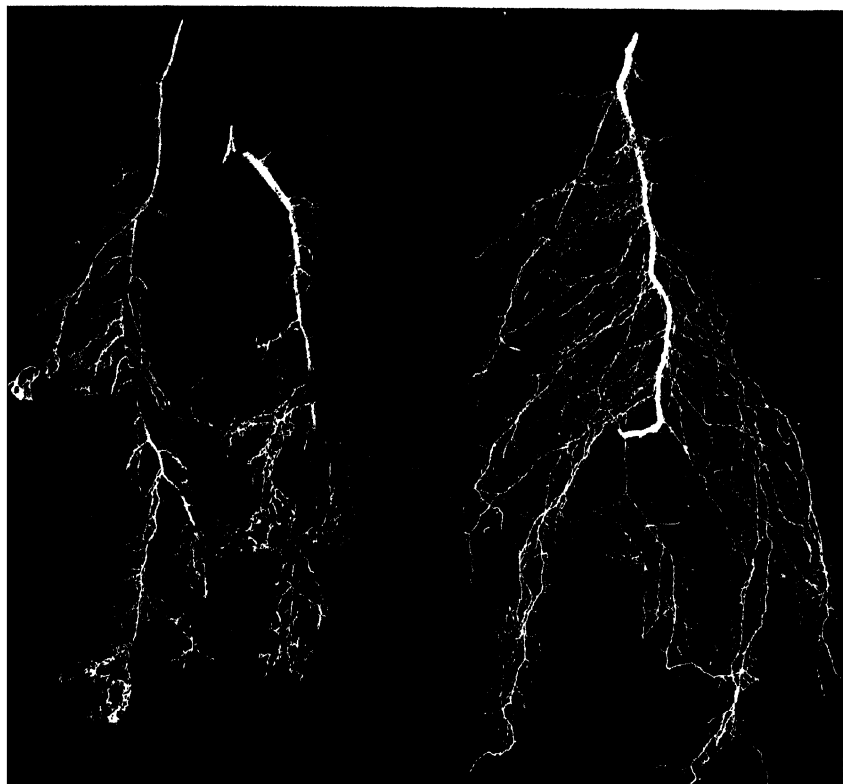


Fig. 1

Fig. 2

### PLATE 8

Fig. 1.—Thickened rootlets and proliferated root tips of corn injured by 0.1 per cent of copper added as copper sulphate to the soil. ( $\times 3$  diam.)

Fig. 2.—Fine roots and root tips of corn, normal. ( $\times 3$  diam.)  
(Photos by G. F. Freeman.)



Fig. 1

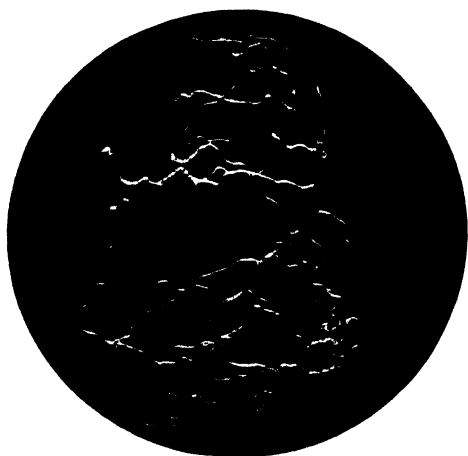
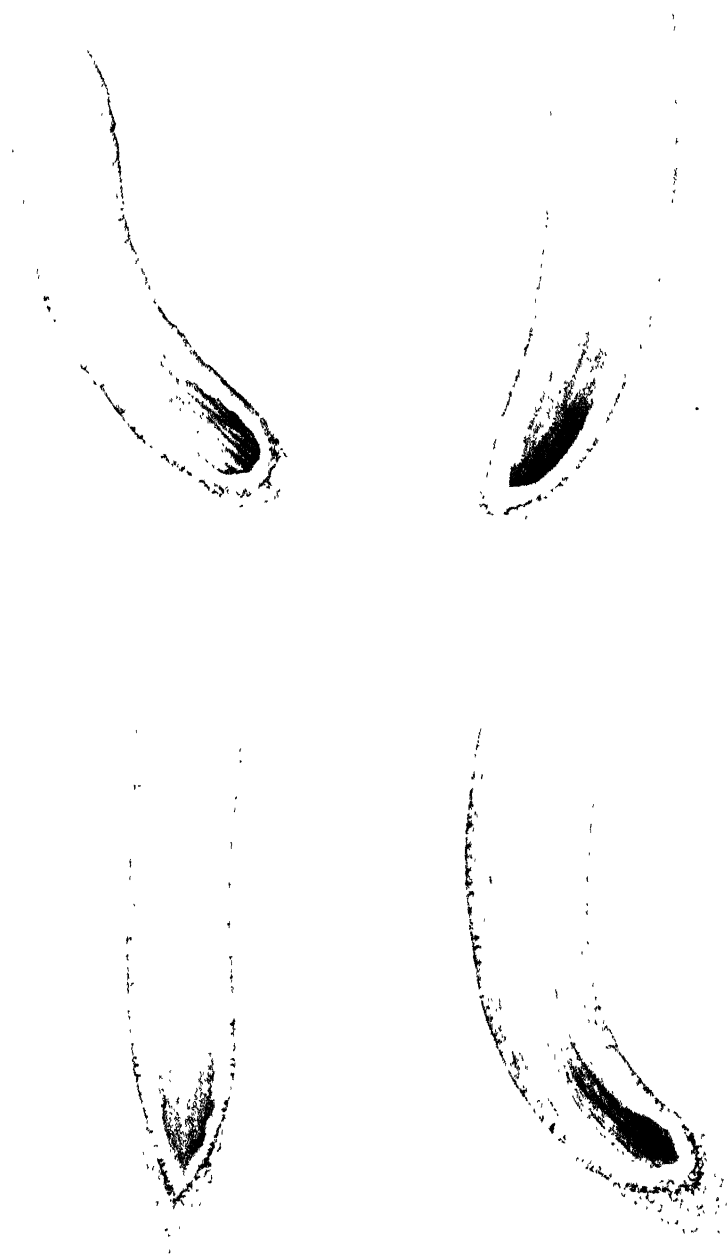


Fig. 2

### PLATE 9

Corn root-tips killed in a solution of 1 part copper to 100,000 of water, and colored by means of (1) caustic potash, which gives the violet biuret reaction, identifying both copper and protein; (2) hydrogen sulphide, brown; (3) potassium xanthate, yellow; and (4) potassium ferrocyanide, red. ( $\times \pm 30$  diam.)







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EXPERIMENTS ON THE EFFECTS OF CON-  
STITUENTS OF SOLID SMELTER  
WASTES ON BARLEY GROWTH  
IN POT CULTURES

BY

C. B. LIPMAN AND W. F. GERICKE

UNIVERSITY OF CALIFORNIA PRESS  
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## INTRODUCTION

In 1913<sup>1</sup> the senior author and F. H. Wilson reported briefly the results of some preliminary investigations on the effects of  $\text{CuSO}_4$ ,  $\text{MnSO}_4$ ,  $\text{ZnSO}_4$ , and  $\text{H}_2\text{SO}_4$  on the growth of wheat and of vetch in a humus sand in pots, under greenhouse conditions. One year prior to the appearance of the report just cited, the present authors instituted new and more complete experiments with the objects noted below. These experiments, covering a period of three years, are now complete in several significant phases and we are therefore proceeding to a discussion of them.

The importance of a study of this subject is attested by the recent appearance of monographic works devoted to it, by the significance of the practical bearings of the physiological studies involved and, in view of these, by the conflicting nature of the results thus far obtained, and the evident non-consideration, by investigators, of the nature of the medium of plant growth as a vital determinant of the results. Some of the outstanding early work on the inorganic poisons, particularly copper, as affecting plant growth is either reviewed or cited in the communication above referred to. In this paper no historical sketch will be given, but important investigations which may be relevant to our findings will be discussed in connection with the results and meaning of our experiments.

## OBJECTS OF THE EXPERIMENTS

The objects of our experiments were as follows: (1) To ascertain whether metals like copper, zinc, lead, iron, and manganese used in the sulfate form in the soil as a medium are toxic in any quantity to barley. (2) To ascertain whether the substances named can be toxic in soils if found in quantities which

<sup>1</sup> Bot. Gaz., vol. 55, no. 6, p. 409, June, 1913.

would be possible in the vicinity of smelters. (3) To ascertain whether the same substances would be a menace to lands more remote from smelters if carried down to them in solution in irrigation water of streams polluted by solid smelter wastes. (4) To ascertain whether the compounds named may exercise a stimulating effect on plants grown in soil as a medium and, if so, whether the effect noted is ephemeral or permanent in one way or another. (5) To ascertain whether potassium aluminum sulfate can have any value as a source of potash or as a plant stimulant.

### METHODS OF THE EXPERIMENTS

The experiments were carried out in the greenhouse, the successive crops being grown at different seasons of the year so as to allow a study of the effects of a variety of climatic conditions. The plants were not artificially shaded during the period of growth. The soil used in most of the experiments was a clay adobe containing a very good supply of organic matter to start with, and was made up by adding barnyard manure to our hillside clay adobe soil. The other soils employed in a number of the experiments which served as checks on the heavier soils were a blow sand from Oakley, California, and the clay adobe soil above named unmixed with manure. Evidence is thus obtained of the effects on barley of at least one of the salts mentioned, in four types of soils, since a humus sand was, as explained above, employed in the preliminary experiments. The chemical analysis by the Hilgard strong acid digestion method of the humus clay adobe, of the blow sand, and of the clay adobe yielded the results shown in table 1 (p. 498).

The containers for the soils just described were ordinary earthenware pots nine inches in diameter at the top. These pots were paraffined to preclude the possibility of the absorption of salts by the porous walls. From ten to twelve pounds of soil were used per pot, depending upon the kind of soil employed. The salts were applied in solution in all cases except in that of the lead sulfate, which, owing to its insolubility, was mixed, in the form of powder, with the soil. The mixing was done as

TABLE I  
CHEMICAL ANALYSES OF SOILS USED

	Greenhouse humus clay adobe	Clay adobe	Oakley sand
Insoluble residue .....	74.03	85.50	92.04
Soluble silica .....	9.18	7.40	3.14
Lime (CaO) .....	2.26	1.05	.66
Iron Oxide (FeSO <sub>4</sub> ) .....	4.59	3.61	3.60
Aluminum oxide (Al <sub>2</sub> O <sub>3</sub> ) .....	5.80	3.85	1.24
Sulfuric acid (SO <sub>3</sub> ) .....	.....	.....	.....
Manganese sesqui-oxide (Mn <sub>2</sub> O <sub>3</sub> ) .....	.13	.13	Trace
Magnesium oxide (MgO) .....	.72	.54	.22
Potash (K <sub>2</sub> O) .....	.62	.25	.30
Soda (Na <sub>2</sub> O) .....	.43	.21	.17
Phosphoric acid (P <sub>2</sub> O <sub>5</sub> ) .....	.48	.20	.16
Moisture and volatile matter .....	11.68	4.70	1.72
Total .....	100.78	100.44	100.32
Nitrogen .....	.31	.12	.03
Humus .....	3.20	1.85	.30
Nitrogen in humus .....	6.75	8.00	11.80

thoroughly as possible to approach closely a uniform distribution of the salt. Obviously such thorough mixing as could be desired was not attained with the PbSO<sub>4</sub>; hence one reason for the irregularity of some of the results obtained therewith. In the case of the copper, lead, manganese and zinc sulfates the treatments were made as parts per million of the dry weight of the soil, whereas the ferrous sulfate was supplied in much larger quantities on the percentage basis. The precise quantities of the salts employed are shown in the tables submitted below, but it is added here, in explanation, that the treatments as indicated there represent aggregate amounts in the case of the copper, zinc, iron, and potash alum series of two separate applications, *one prior to planting the first and the other prior to planting the second crop* of barley in the humus clay adobe soil. It will further be noted that all the salts were added in the form of sulfates of the metals studied, except as otherwise stated.

Water was applied to the surface of the soil in irrigating. As a rule, that operation was carried out twice a week, or as needed, and 400 c.c. of tap water was the amount used at every irrigation. From earlier tests it appeared that this quantity of

irrigation water and the mode of its application were most desirable under the circumstances and were such as to preclude losses of water and salts by percolation and drainage.

Barley (*Hordeum vulgare*) was the crop grown throughout all series of experiments. The variety employed was a selected and vigorous strain of Beldi. Three crops were grown in succession on the humus clay adobe soil, the first and third crops being produced in the period between September and January of the years 1912-1913 and 1913-1914 respectively, and the second crop between March and June, 1913. Only two crops were grown on the non-humus clay adobe soil, in periods corresponding to the last two for the humus clay adobe soil. One crop only was grown on the blow-sand soil.

At the time of harvest, which was carried out when the grain was thoroughly mature, the plants were cut as close as possible to the ground. The total crop thus obtained was placed in paper bags and dried until the weight was constant. Then the weights, separately, of grain and straw were determined. At the same time the soil in the pots was thoroughly worked over to obtain the roots produced in every case. In some instances nitrification studies on the soil were made, and also determinations of the amounts of salts remaining behind in the soils after harvest, and the amounts taken up by the crop. Enough of these analyses, as well as of nitrogen determinations in the grain, were accomplished to ascertain the tendency of these conditions in the plants and soils studied.

## RESULTS OF THE EXPERIMENTS

It will undoubtedly be of interest to our readers to learn first, from the results of our experiments, something of the appearance, height, tillering, color, and similar observations on the growth of the barley, and later the yields obtained, composition of the grain, and changes in the soil. The following general statements may therefore be made at this point with respect to the first class of data obtained through the experiments. The different features will be considered separately.

## COLOR OF LEAVES

In the first crop on the greenhouse soil the color of the blade was a much deeper green in the treated pots, no matter what salt was used, than in the untreated ones. This was true despite the fact that the leaves were dark green in the plants of the control pots, which had a plentiful supply of available nitrogen at their disposal. The plants of the pots treated with copper sulfate showed, however, a darker green color than those in the pots treated with other salts. That excessive nitrogen feeding was probably the cause of the very deep green color of the leaves referred to was further indicated by the tendency to lodge manifested by the plants in the copper and lead series and to some extent in the other series. In the second crop, prior to the planting of which the salt content of all series in the greenhouse soil, except the lead and the manganese, was doubled, manifestations as to color similar to those above described were observed. These were not so marked, however, even though the contrast between the plants on the treated and those on the untreated soils was easily discernible. As a result of the smaller amount of stimulation in the crop under consideration, no tendency to lodging was noted, and the plants were erect and rigid. In the third crop on the same soil without further salt treatment, there was only a slightly deeper green color in the leaves of plants on the treated than in those on the untreated soil. Again, the plants were erect and vigorous in appearance throughout.

On the Oakley blow-sand soil, in which only copper was tested, was discerned a similarly striking effect on the color of the barley blade exercised by the salt treatment of the soil. In the clay adobe soil similar observations were made. We are therefore led to believe that the effect of the salts in question seems to be general in at least one direction for all soils, namely, for the production of a deeper green color in the leaves of plants growing on the treated soil. The stimulating effect in that direction shows a tendency to diminish at first rapidly, then slowly, in the succeeding crops. The probable causes of this manifestation, as briefly referred to above, will be mentioned later in connection with the studies of the treated soils them-



selves. As further evidence of the general nature of the effects of the salts in question in soils on the color of the leaves of plants, we may cite again the observations on that point made by both Pammel<sup>2</sup> and Van Slyke.<sup>3</sup> Those investigators reported marked deepening of the color of the leaves of tomatoes and other plants due to treatment of the culture soil with  $\text{CuSO}_4$ . Other kinds of plants, therefore, as well as other soil types than those employed by us seem to be similarly influenced by  $\text{CuSO}_4$  with reference to color production in leaves.

#### TILLERING

During the first two crops grown, the amount of tillering occurring in the plants on the greenhouse soil was studied. This was done with the idea of noting if any close correlation existed between the amount of growth and dry matter produced by the treatment and the number of tillers formed. Our observations give a negative reply to this query. Thus in the first crop of the copper series the number of tillers per pot of four plants varied from thirteen to thirty-one over the whole range of concentrations of copper sulfate employed. This in itself would of course be of little significance in connection with the question under consideration if there was a decrease or an increase in the number of tillers with a change in concentration of  $\text{CuSO}_4$ . This was not strictly the case, however, and to illustrate we may say that the largest number of tillers in the first crop of the  $\text{CuSO}_4$  series was in one of the pots receiving 1500 p. p. m.  $\text{CuSO}_4$ ; the smallest number of tillers was produced in the pots remaining untreated. Moreover, there was but little agreement in that respect between duplicate pots receiving  $\text{CuSO}_4$ . Thus the duplicate of the pot above mentioned as producing the largest number of tillers (thirty-one) produced only twenty-one, and such large discrepancies between duplicate pots were common. The fact remains, nevertheless, that while small concentrations and large concentrations of  $\text{CuSO}_4$  do not differ in their effects on the number of tillers, some  $\text{CuSO}_4$  as against no  $\text{CuSO}_4$  appears to be of definite effect in the first crop. Thus in the large

<sup>2</sup> Iowa Agr. Exp. Sta. Bull. no. 16, 1892.

<sup>3</sup> N. Y. (Geneva) Agr. Exp. Sta. Bull. no. 41, 1892.

range of concentrations employed in the copper series, there was no pot receiving  $\text{CuSO}_4$  in any quantity which did not produce more tillers than any of the control pots, which showed from thirteen to fourteen tillers in each of three pots employed as untreated controls. In general, therefore, it seems that in the first crop, copper sulfate does stimulate tillering, but it does so irregularly and small amounts of the salt appear to be as effective in that direction as large amounts. In the second crop of the  $\text{CuSO}_4$  series the number of tillers was decreased throughout because of climate and other obvious effects accompanying the conditions of the experiment which are above described. Nevertheless, the treated pots were, with very few exceptions, decidedly superior in tiller production to the untreated pots, which, again, agreed well among themselves. Otherwise, the tillering of the second crop in the  $\text{CuSO}_4$  series was not significantly different from that of the first crop.

In the first crop of the zinc sulfate series the number of tillers was very markedly larger than in the first crop of the copper sulfate series. Thus the largest number of tillers produced in the first crop of the copper series is about equivalent to the smallest number of tillers in the first crop of the zinc sulfate series. In one pot of the zinc series sixty-five tillers were produced by four plants, a number more than twice as great as the maximum in the first crop of the copper series. Moreover, the agreement between duplicate pots was on the whole much better in this regard in the zinc than in the copper series, despite the fact that several large discrepancies were noted. In the first crop, therefore, the zinc sulfate, like copper sulfate, has not only stimulated tiller production, but has done so to a much more marked degree than the last-named substance. In the second crop, however, conditions and results are considerably changed. Thus, whereas the stimulating effects of  $\text{CuSO}_4$  on tiller production are clearly manifest throughout the series, even after the salt concentration is doubled in the same soil, only one case of stimulation in that respect (at the lowest concentration) is noted in the  $\text{ZnSO}_4$  series under similar conditions. Moreover, in the  $\text{CuSO}_4$  series we find scarcely one undoubted case of depression in tillering even in the second crop, due to

the treatment of the soil, but such depression obtains almost without exception in the second crop of the  $\text{ZnSO}_4$  series. In other words, a very marked decrease in the number of tillers results from the second  $\text{ZnSO}_4$  application to the greenhouse soil, both absolutely and relatively speaking, in comparison with either the untreated control pots or with the treated pots of the copper series. Much better agreement between duplicate pots with reference to tillering is noted in the second crop of the  $\text{ZnSO}_4$  series than in the other series above described.

In the case of the potash alum series, in the first crop the results were very similar to those obtained in the corresponding copper series except that tillering was not so markedly stimulated as in the latter. In the second crop, also, the results of the potash alum series were not strikingly different as regards tillering from those in the copper series.

In the first crop of the  $\text{FeSO}_4$  and  $\text{PbSO}_4$  series no observations were made on tillering owing to the poor development of plants and their prostrate mode of growth, which was especially marked in the lead series. Observations on the amount of tillering produced in the second crop of the  $\text{FeSO}_4$  series indicated that the stimulating effect of the  $\text{FeSO}_4$  on tillering was not so great as that of either copper or potash alum, but greater than that of zinc.

In the third crops of all series scarcely any tillering was observed, the plants producing for the most part single upright stalks. It appears, therefore, that the stimulating effects of the salts tested with respect to tillering are ephemeral in their nature, but are more distinctly so with some salts than with others.

#### HEIGHT OF PLANTS

In the second crop only, observations and measurements were made of the average and total heights of plants produced in the  $\text{CuSO}_4$ ,  $\text{ZnSO}_4$ ,  $\text{FeSO}_4$ , and potash alum series. These indicated definite increases in height of plants produced by certain concentrations of all the salts named, over those attained by the plants in the untreated pots. The superior height of the plants was, however, variously distributed through the series. Thus it was

apparent in the first five lowest concentrations of the  $\text{CuSO}_4$  series. It did not show in the first two concentrations of  $\text{ZnSO}_4$ , but in all others, and was clearly manifest almost throughout the potash alum series. In the  $\text{FeSO}_4$  series the heights followed the general observations just recorded for the  $\text{ZnSO}_4$  series so far as comparison with controls is concerned. As regards absolute heights of plants the  $\text{ZnSO}_4$  series showed the highest, and the potash alum series was a close second, with the others considerably behind.

It appears, therefore, that with regard to stimulation both of tillering and of tallness,  $\text{ZnSO}_4$  is superior to the other salts. The agreement between duplicate pots in respect to the height of plants was far more satisfactory than that for tillering. None of the actual data are given here because of the necessity for brevity in such papers and because of the decidedly minor significance of the results in connection with the main issue under examination.

#### GERMINATION OF SEEDS

In all of the series under discussion, the germination of seeds was more rapid in the salt-treated soils than in those untreated, at least up to certain very considerable concentrations of the salts. Exceptions to this rule were of course found in certain concentrations of the salts which entirely inhibited growth and in those which almost did so; but in all pots in which the salt concentrations were not of that order, germination was much more rapid than in those which remained untreated. The stimulation in respect to germination was about of the same degree in all concentrations of every salt which would at all stimulate germination, larger amounts of salts not differing from the smaller ones. Certain definite differences, however, existed in that regard among the different salts. Thus  $\text{CuSO}_4$  stimulated germination most,  $\text{ZnSO}_4$  was second in order,  $\text{FeSO}_4$  third, and the other salts exerted only a slight influence. Our findings in this respect, therefore, are again in accord with those of Pammel<sup>4</sup> and Van Slyke,<sup>5</sup> which are above cited, and also with

<sup>4</sup> Iowa Agr. Exp. Sta. Bull. no. 16, 1892.

<sup>5</sup> N. Y. (Geneva) Agr. Exp. Sta. Bull. no. 41, 1892.

those of many other investigators, among whom may be mentioned Effront.<sup>6</sup> It may be added here that the stimulating effects of the salts studied with respect to germination of seed were noted in the first and second crops. In the third crop there was little, if any, superiority in germination of the seeds in the treated as against the untreated pots. In other words, we have noted that in regard to germination, as well as in respect to tillering and other superficial characters, the salts employed stimulated the barley for one or two seasons at certain concentrations and after that showed no marked effect in either direction. It should also be observed that in cases in which such salts as  $\text{CuSO}_4$  at higher concentrations retarded germination in the first crop, the retarding effect disappeared in the second and third crops.

### YIELDS OBTAINED

In studying the yields of barley in all the series, the weights of straw, of grain, and of roots were determined in every case after drying at  $100^\circ \text{C}$  and bringing to constant weight. All such determinations are given in the tables which follow, together with other necessary data. It will be noted that the yields of the single pots in every duplicate pair are given, as well as the averages. This is for the purpose of pointing out the large variations in yield frequently obtained in duplicate pots of soil cultures and for that of allowing our colleagues to study our data at first hand and reach their own conclusions. The different salts will be considered separately below, with one type of soil at a time.

#### COPPER SULFATE—GREENHOUSE SOIL

Tables IIa, IIb, and IIc give the results obtained with  $\text{CuSO}_4$  in three successive crops on the greenhouse soil. Through an error, as was stated above, a second application of  $\text{CuSO}_4$  equivalent to the first was made to the soil prior to planting the second crop, so that for the second and third crops, amounts of  $\text{CuSO}_4$  were present in the soil which were far larger than those intended for the study. While, therefore, we have obtained

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<sup>6</sup> Effront, J., *Comptes Rendus Acad. Sci. (Paris)*, vol. 141, p. 625, 1905.

three successive crops on the same soil, only one of them, the third, was influenced purely by the residual effects of the  $\text{CuSO}_4$  application remaining after the production of one crop. With these observations in mind, let us now consider the results of the  $\text{CuSO}_4$  applications in the three crops harvested on the greenhouse soil.

#### FIRST CROP

The growth of the plants in the first crop was very rank in the controls as well as in the treated pots, with the result that high yields of dry matter were obtained. This was doubtless due to a large supply of available plant food in the fresh greenhouse soil and especially to the high nitrate content and high nitrifying power of the soil. The deep green color of the leaves, above referred to and the tendency shown by the plants to lodge seem to confirm this view, and it is further supported by nitrification studies which we carried out and which are reported below. Because of the conditions for rank growth, however, the growing season was lengthened and scarcely any grain was produced. The data of the first section of table II therefore give only the yields of straw and roots. Despite considerable discrepancy among the yields of duplicate pots, there can be no question after an examination of the data for the first crop that  $\text{CuSO}_4$ , in the concentrations and under the conditions employed, has caused the barley to produce more dry matter than was produced in the control pots. Such stimulated growth is apparent throughout the whole series of copper concentrations varying from 50 p. p. m. to 1500 p. p. m. Concentrations above 1000 p. p. m. seem to be definitely more toxic, or at least less stimulating, to the barley plants than lower concentrations if average yields are adopted as criteria. Such procedure may, however, be unjustifiable because of the large discrepancies among the yields of duplicate pots. That the increases in yields of dry matter of barley are real and not accidental is evidenced not only by their manifestation in the whole series, but also by the magnitude of the increases involved. Thus in the concentration of 600 p. p. m.  $\text{CuSO}_4$  an increase in yield over that of the control pot was obtained which was equivalent to nearly 50 per cent of the total

yield of the latter, and in several other cases such increases amounted to 30 per cent or 35 per cent.

Giving brief consideration now to the yields of roots alone, we find that they, too, like the total dry matter in general, are definitely affected by the  $\text{CuSO}_4$  treatment. Increased root production over that in the control pots is found in all the pots having concentrations from 50 p. p. m. to 600 p. p. m.  $\text{CuSO}_4$ , inclusive. Beyond that point, however, unlike the case of the total dry matter considered, the increased concentrations of  $\text{CuSO}_4$  appear to depress root development very definitely. The decreases continue steadily more significant as the concentration of  $\text{CuSO}_4$  increases from 600 p. p. m. to 1100 p. p. m., when the toxic effect seems to reach a stationary point and no further decreases occur, even though more  $\text{CuSO}_4$  is added up to concentrations of 1500 p. p. m.

Taking into consideration the effects of  $\text{CuSO}_4$  on the first crop of barley in the greenhouse soil in regard to both tops and roots produced, it appears that we must consider the point of stimulation to cease at 600 p. p. m.  $\text{CuSO}_4$ . It is possible in addition that even the 700 and 800 p. p. m. concentrations may be looked upon as still stimulating to both tops and roots of the barley plant in the soil in question. Beyond those points, however,  $\text{CuSO}_4$  is stimulating, in the first crop, to the production of tops only, not to the production of roots.

#### SECOND CROP

The very large decrease in yield of the second crop in the same pots, so far as total dry matter is concerned, is clearly indicated in table IIb. This is evidently not due to the doubling of the percentage of  $\text{CuSO}_4$  in pots receiving treatment, since the decrease in the second as compared with the first crop is just as clearly marked in the control pots. On the other hand, whereas the first crop produced practically no grain, probably for reasons above discussed, the second crop produced a large yield of grain, amounting not infrequently to 25 per cent or considerably more of the total dry matter. Again, we see in the figures for the second crop the disparity between yields of duplicates, but again also the consistently large yields of dry

matter in the treated as against those of the untreated pots. This is strikingly so for both the straw and the grain yield, but is most consistently and undeniably apparent in the latter. The root yields in most of the treated pots are also superior to those obtained in the untreated pots, and duplicate cultures show better agreement in that respect than do the straw and the grain yields. The grain produced was in all cases well filled and normal in appearance. In brief, we find that the second crop on the soil treated with  $\text{CuSO}_4$ , despite the doubling of the  $\text{CuSO}_4$  application, shows as markedly, and perhaps even more markedly, the stimulating effect of the salt under consideration to barley grown on greenhouse soil. While in detail the results of the second crop differ from those of the first crop, they appear to confirm the latter in general. The average yields of dry matter are greater with all treatments than they are in the untreated pots. This strikingly stimulating effect of  $\text{CuSO}_4$  on barley under the conditions named in concentrations reaching a maximum of 0.3 per cent  $\text{CuSO}_4$ , based on the dry weight of the soil, is as astounding as it is interesting, and it would appear to lend little support to the idea of the toxicity of  $\text{CuSO}_4$  in relatively small amounts to crops grown on field soils. This phase of the subject will, however, receive more attention below.

### THIRD CROP

Grown under more propitious weather conditions, as explained above, the third crop in the  $\text{CuSO}_4$  series on the greenhouse soil yielded throughout much larger amounts of dry matter than the second crop, though not such large amounts as the first crop. Again, we note the general stimulating effect of  $\text{CuSO}_4$  to the production of dry matter in barley plants. This time, it should be observed, the stimulating effect was not manifest throughout the treated portion of the series, as it was in the first two crops. Thus four of the  $\text{CuSO}_4$  concentrations employed, namely, 600 p. p. m., 1600 p. p. m., 2400 p. p. m., and 2600 p. p. m., depressed the yield of barley if average yields of duplicate pots are considered. In most cases, however, such depression of yield is easily within the experimental error and therefore may be without significance. This is especially so



since there is no regularity in the inhibiting power of  $\text{CuSO}_4$ , referred to; but small as well as large concentrations at isolated points in the series depressed the yields as above pointed out, whereas the rest of the concentrations, also small and large, stimulated the yields.

While the total dry matter produced in the third crop, as shown above, is greater than that yielded in the second crop, the yield of grain in the latter is far superior to that in the third crop. Thus the highest grain yield in the third crop is scarcely more than one-third that of the second crop, and the lowest yield of the third crop is about one-sixth that of the second crop. Nevertheless these facts are of no significance in connection with the effects of  $\text{CuSO}_4$ , since the control pots manifest the same depression in grain yields which is characteristic of the treated pots in the third crop. Likewise, in most cases the treated pots produced more grain than the untreated pots.

The point of maximum stimulation of  $\text{CuSO}_4$  to the production of dry matter by the barley plant on the greenhouse soil is very difficult to discern. While apparently it occurs at the concentration of from 0.18 per cent to 0.2 per cent  $\text{CuSO}_4$  of the dry weight of the soil, the irregularity and non-agreement of many of the duplicate pots render decisions in such matters unsafe, if not valueless. In general, however, the figures in table IIc leave little room to doubt the non-injurious nature and perhaps the stimulating effect of  $\text{CuSO}_4$ , at considerable concentrations, for barley in the greenhouse soil under the conditions described.

#### COPPER SULFATE—ADOBE SOIL

Tables IIIa and b give the results obtained with  $\text{CuSO}_4$  treatment in the case of the adobe soil in the first and second crops respectively. It will be noted at the outset that the yields on the adobe soil are much lighter than those on the greenhouse soil. The reasons for this circumstance are of course not far to seek, in the light of the origin and descriptions of the soils used in these experiments which are given above. Only two crops were grown on the adobe soil, because we did not decide

to start it for comparison with the greenhouse soil until one crop had been obtained with the latter.

#### FIRST CROP

We find in the case of the adobe soil the same unfortunate disparity among the yields of duplicate pots which was noted with the greenhouse soil. This disparity is of course the more noticeable when much smaller absolute amounts are involved, as in the present case. Despite all that, however, there appears to be justification for the conclusion, based on the data in table IIIa, that  $\text{CuSO}_4$  does exercise a stimulating action on the growth of barley in adobe soil. Such stimulation is not apparent throughout the whole series, as it is in the case of the greenhouse soil, but it appears to exist in all concentrations of  $\text{CuSO}_4$ , employed up to 900 p. p. m. Concentrations in excess of the latter seem to depress, definitely, the yield on the adobe soil. But whether or not we admit the existence of a stimulating effect by  $\text{CuSO}_4$  on the barley, based on the figures here studied, it can scarcely be denied that  $\text{CuSO}_4$  is not toxic to barley in the first crop grown on adobe soil until nearly 0.1 per cent  $\text{CuSO}_4$  is present in the soil. Amounts of  $\text{CuSO}_4$  slightly less than 0.1 per cent stimulate the growth of the barley significantly. At concentrations of 0.15 per cent and 0.2 per cent  $\text{CuSO}_4$  no growth is obtained at all, showing of course marked toxicity.

Some interesting facts are brought to light in table IIIa with respect to the relationships among straw, grain, and root yields which obtain between treated and untreated soils and among themselves in the case of the different concentrations of the latter. In the first place, it will be noted that the grain yields form an even larger percentage of the total dry matter of the first crop on the adobe soil than they do in the second crop on the greenhouse soil. In some cases, indeed, the average yield of grain in duplicate pots exceeded the average yield of straw. In nearly half the treatments, the grain yields were larger than those of the control pots so that the stimulating effect of the  $\text{CuSO}_4$  application, if allowed, applies to the grain yields as well as to those of total dry matter. The root yields are pro-

portionately smaller on the adobe than on the greenhouse soil, as are the yields of the barley as to tops. Nevertheless, the average yields of roots also show the stimulating effects of  $\text{CuSO}_4$ , since they are greater in all concentrations than those of the control pots until a concentration of 1000 p. p. m.  $\text{CuSO}_4$ , or 0.1 per cent based on the dry weight of the soil, is reached. In excess of that concentration,  $\text{CuSO}_4$  is toxic to roots and appears to inhibit their development.

#### SECOND CROP

Much better agreement among the yields of straw in duplicate pots of the second crop on the adobe soil was obtained than in any of the series with  $\text{CuSO}_4$  above described. In fact, the agreement between the duplicates in nearly all cases was as good as could possibly be hoped for when one allows for the ever-present idiosyncrasies of plant protoplasm. Up to and including concentrations of 400 p. p. m.,  $\text{CuSO}_4$  seemed to depress barley growth except in one concentration, namely, at 300 p. p. m. Such depression is probably not significant, except at the concentration of 100 p. p. m. However that may be,  $\text{CuSO}_4$  did not stimulate the development of barley at the lower concentrations in the second crop on the adobe soil as it did, with one exception, in the first crop. On the contrary, conditions reversed themselves in the second crop, and the most marked and consistent stimulation occurred in the higher concentrations of  $\text{CuSO}_4$ , only the very highest concentration—namely, 2000 p. p. m.—showing a more or less definitely toxic effect. Thus, while no growth was obtained in the first crop on the adobe soil containing 1500 p. p. m. of  $\text{CuSO}_4$ , the same soil on the second planting stimulated the growth of barley so that in both pots, taken separately and by averages, the yield was superior to that of the control pots. Irregularities of course crept into this series as into the others, for instance, depressed growth or no stimulation at a concentration of 1200 p. p. m., when stimulated growth is obtained at 100 p. p. m.  $\text{CuSO}_4$  on the one hand and at 1500 p. p. m. on the other, is a circumstance which is very difficult of explanation.

Again, unlike the first crop on the adobe soil, the second crop yielded no grain. This of course cannot be attributed in any way to the effects of  $\text{CuSO}_4$ , since the control pots behaved in the respect noted like the treated ones. Presumably, unfavorable climatic conditions and the heavy nature of the soil may have produced and influenced the result obtained. The root yields, however, were very considerably larger in the second than in the first crop, and fairly good agreement between duplicate determinations was obtained. Considering the small total yield of dry matter in roots, it is perhaps a very significant stimulation to their development which  $\text{CuSO}_4$  exerts.

Regarded then from the standpoint of the total dry matter produced, there appears to be no question from the data in table IIIb that  $\text{CuSO}_4$  can stimulate growth in the barley plant on a clay adobe soil even when present at very considerable concentrations. If only the dry matter of the above-ground parts is considered, three exceptions to this rule in the whole series can be found. In a general way, the results with  $\text{CuSO}_4$  on the adobe soil confirm those obtained on the greenhouse soil using the same salt. It may be repeated with advantage here that even if such stimulating properties of  $\text{CuSO}_4$ , which in our opinion we have shown above to exist, are not allowed, our data do not offer any support to the idea that in the ordinary quantities in which copper may be introduced in agricultural soils it is even likely to be toxic to grain plants.

#### COPPER SULFATE—OAKLEY SOIL

Only one crop of barley was grown on the Oakley soil in the tests with  $\text{CuSO}_4$ . Since closing the experiment we have regretted the fact that the Oakley soil was not cropped successively for two or three seasons after treatment, but at the time of the experiment this was not deemed necessary. Table IV gives the results obtained with the one crop in question. The figures really do not tell the whole story, since the appearance of the barley plants was far superior on the treated soils on which they developed at all than it was on the untreated soil. Nevertheless, the figures are striking enough to be used alone

as a criterion to determine the effects of  $\text{CuSO}_4$  on the growth of barley in the Oakley sand. Our data show very clearly the stimulating effect of  $\text{CuSO}_4$  for barley in the first crop on the Oakley sand. They also show, more clearly than any series above described, the toxic effect exercised by  $\text{CuSO}_4$  at the higher concentrations. The stimulating effects further do not occur at such high concentrations of  $\text{CuSO}_4$  in the case of the Oakley sand as in that of the greenhouse soil or even in that of the adobe soil. To be specific, we find that at concentrations of 100, 200, and 300 p. p. m.,  $\text{CuSO}_4$  is definitely stimulating to barley production on the Oakley soil under the conditions of our experiment. The most marked results of the stimulation in question are not manifest in the production of straw or even in that of roots when it is at all perceptible, but is very marked in nearly all cases so far as grain production is concerned. It is a curious fact that at a concentration of 600 p. p. m.  $\text{CuSO}_4$  in the Oakley soil, we obtain the largest grain production of the whole series, and yet the straw production is depressed through the  $\text{CuSO}_4$  treatment at that concentration and the root development almost entirely inhibited. This fact is very difficult to explain, but exhibits parallelism to similar facts observed by both Pammel and Van Slyke in the experiments above cited. When the dry matter produced is considered as a whole, and straw, grain, and roots are considered together, stimulation is noted only in the case of the first two concentrations of  $\text{CuSO}_4$  employed, and the stimulation is not very marked. In other words, one is obliged to state definitely the criterion employed when forming a judgment as to the existence or non-existence of a stimulating effect of  $\text{CuSO}_4$  on barley grown on the Oakley sand. It will be necessary in the future, for a more decided judgment of the question in hand, to grow several successive crops of barley on the soil named, once treated with  $\text{CuSO}_4$ , as shown in the table, and possibly also to supply available nitrogen, which is a serious limiting factor in the growth of barley on that soil. Without making any final statements in the premises, however, the data given by us in table IV seem to point strongly to the existence of a stimulating action of  $\text{CuSO}_4$  to barley growth, even on the Oakley sand.

## ZINC SULFATE—GREENHOUSE SOIL

Only the greenhouse soil was employed to test the effect of  $\text{ZnSO}_4$  on barley plants. As was the case with  $\text{CuSO}_4$  on the same soil, three successive crops were grown, two treatments of  $\text{ZnSO}_4$  being given. The results obtained, together with the treatments given, are indicated in tables Va, Vb, and Vc.

## FIRST CROP

A study of table Va reveals the fact that  $\text{ZnSO}_4$  in the case of the first crop of barley is not unlike  $\text{CuSO}_4$  in its action. In other words, while the latter salt exercises a greater and more definite stimulating action in the first crop,  $\text{ZnSO}_4$  also manifests a definite though smaller stimulating effect on the barley plants. This seems to be supported by the fact that only eight pots out of thirty treated with varying amounts of  $\text{ZnSO}_4$  give a smaller yield than the highest yield of the control pots. In general, the stimulation seems to be greatest at concentrations of  $\text{ZnSO}_4$  varying from 500 p. p. m. to 1200 p. p. m. inclusive. This statement has reference only to the total yield of dry matter and not to any parts, like roots or tops, taken separately. On the same bases, also, no definitely toxic effect of  $\text{ZnSO}_4$  was observed, though, as above intimated, some apparent effects of that nature were noted. Again, in accord with the results obtained with  $\text{CuSO}_4$  no grain worth weighing was produced in the first crop, and the weights of the straw given in the table therefore include such partially formed heads as were developed. In still further agreement with the results of the first crop of the  $\text{CuSO}_4$  series,  $\text{ZnSO}_4$  stimulated the growth and development of roots practically throughout the whole series. The stimulation to root development alone was, however, greater in the  $\text{ZnSO}_4$  series than in the  $\text{CuSO}_4$  series, just as the opposite was true for the tops. The greatest stimulation to root development appears to have been attained at the higher rather than at the lower concentrations of  $\text{ZnSO}_4$ , the difference being most marked in that respect between the first three concentrations employed and the rest. This circumstance, as will be seen by a comparison of table Va with table IIa, is the reverse of that noted in the first crop of the  $\text{CuSO}_4$  series, in which the first four

concentrations gave the largest increases of dry matter of roots over the controls. All in all, the effect of  $\text{ZnSO}_4$  in the case of the first crop on the greenhouse soil must be regarded as one definitely stimulating to the production of dry matter in the barley plant.

#### SECOND CROP

While in the first crop the  $\text{CuSO}_4$  and  $\text{ZnSO}_4$  series are in general similar so far as the effects of the salts on the barley plants are concerned, they differ markedly in the second crop. To illustrate, it may first be noted in table Vb herewith, on the basis of the total dry matter produced, that  $\text{ZnSO}_4$  beyond concentrations of 600 p. p. m. is distinctly toxic to barley in the greenhouse soil. With similar concentrations of  $\text{CuSO}_4$  in the second crop, the latter salt was not only not toxic beyond 600 p. p. m., but was actually more stimulating at most of the higher than at the lower concentrations. It would therefore seem that so far as the yields of the total dry matter are concerned,  $\text{ZnSO}_4$  is either more toxic than  $\text{CuSO}_4$  or the latter is more readily adsorbed by the greenhouse soil and thus removed from the active solution which bathes the feeding roots. It must, nevertheless, be added that while  $\text{ZnSO}_4$  appears to be definitely more toxic to barley than  $\text{CuSO}_4$  in the greenhouse soil, it cannot be considered very toxic since 0.06 per cent  $\text{ZnSO}_4$  of the dry weight of the soil is not only not toxic, but actually stimulating. We may now consider for a moment the different components of the total dry matter produced in the second crop of the  $\text{ZnSO}_4$  series. So far as the straw alone is concerned, only a concentration of 200 p. p. m.  $\text{ZnSO}_4$  gave stimulating effects. That concentration produced a very marked stimulation, and good agreement is evident in the duplicate pots. Concentrations in excess of 200 p. p. m. depress straw production. Such depression, however, is in some instances not very great, and considerable disagreement between duplicates here, as in the copper series, renders it difficult to pass final judgment in the matter. In general, there is little difference in the depressing effects on straw production of concentrations of  $\text{ZnSO}_4$  varying between 600 p. p. m. and 3000 p. p. m. Beyond 3000 p. p. m. a more definite depress-

ing effect on the production of straw in the second crop becomes apparent. The fact that the wide range of concentrations just referred to is productive of similar effects seems to indicate that most of  $\text{ZnSO}_4$  is adsorbed by the soil and but little of it is free to affect the plant in the soil solution.

With the exception of one or two doubtful cases, grain production is somewhat depressed throughout the second crop of the  $\text{ZnSO}_4$  series. This appears to be even more true for the first concentration of  $\text{ZnSO}_4$ , which stimulates straw production, than for the higher concentrations, which depress straw production. All of these judgments, however, are based on averages of duplicate pots which do not agree very well, and hence considerable caution is employed in stating them. Again, the effect of  $\text{ZnSO}_4$  on grain production seems to be about the same whether small or large quantities of the salt are employed.

So far as root production is concerned, however, the data of the second crop in the  $\text{ZnSO}_4$  series are very different from those bearing on the yields of grain and straw. With three exceptions, two of them at the highest concentrations of  $\text{ZnSO}_4$  employed, the latter induced the production in all pots of more roots than were produced in the control pots. While in many cases the stimulation in the direction noted was not great, it was definite, and in many other cases it was very considerable. Moreover, there was good agreement between the duplicate determinations. The greatest stimulation to root production occurred between 200 and 2600 p. p. m.  $\text{ZnSO}_4$ . While, therefore, the second crop of the  $\text{ZnSO}_4$  series differed from that of the  $\text{CuSO}_4$  series with respect to grain production, there was great similarity in action between the two as regards root yields.

#### THIRD CROP

In the third crop of the  $\text{ZnSO}_4$  series, the toxicity of  $\text{ZnSO}_4$  appears to have become augmented even over that of the second crop. There seems to be no case of stimulation even in the lowest concentration (200 p. p. m.). This applies to straw, grain, and roots equally well. On the other hand, the toxicity of  $\text{ZnSO}_4$  for root and grain production by barley in the third crop is certainly not very marked, although uniform. For straw



production,  $\text{ZnSO}_4$  becomes suddenly very much more toxic than at the lower concentrations when more than 2400 p. p. m. is employed in the culture medium here used. At 200 p. p. m. neither the straw and grain on the one hand, nor the roots on the other, are seriously affected in one direction or another by the  $\text{ZnSO}_4$  in the third crop. Beyond these remarks no discussion of table Vc is necessary. The figures given in that table speak plainly enough for themselves. In contrast with the  $\text{CuSO}_4$  series of the third crop, however, table Vc shows  $\text{ZnSO}_4$  to be again totally different in its effect on the barley plant in the greenhouse soil. Thus the  $\text{CuSO}_4$  exercises a stimulating effect on total dry-matter production in the third crop at almost all concentrations, while  $\text{ZnSO}_4$ , far from doing so in any case, is actually toxic in all concentrations in the third crop on the same soil. This points clearly to sharp differences in the specific physiological effects of the copper and zinc ions, since they were the only apparent variables in question in this experiment.

#### FERROUS SULFATE—GREENHOUSE SOIL

Owing to the rapidity with which ferrous salts are rendered insoluble in well-aerated soils, it was deemed advisable to depart from the procedure followed in the other series so far as quantity of  $\text{FeSO}_4$  used is concerned. Applications of the salt were therefore made at intervals of 0.1 per cent between two succeeding cultures in the series, the lowest concentration used in the first crop being 0.1 per cent, and the highest 1 per cent. As in the other series above described on the greenhouse soil (the only one used in the  $\text{FeSO}_4$  series), the amounts of salts were doubled prior to planting the second crop. The results of the yields are given in tables VIa, VIb, and VIc.

#### FIRST CROP

Even though the amounts of  $\text{FeSO}_4$  used were very large, we see clearly from table VIa that the salt stimulated, in the first crop, the production of dry matter in barley. Considering averages of duplicate pots, we find that there is no concentration

of  $\text{FeSO}_4$  which did not yield increased growth of barley. This constitutes the most striking set of stimulations noted in the series thus far discussed. Moreover, we find again an evident lack of relationship between the amount of  $\text{FeSO}_4$  employed and the degree of stimulation induced thereby. In agreement with the results obtained in the  $\text{CuSO}_4$  and  $\text{ZnSO}_4$  series, the  $\text{FeSO}_4$  series yielded no grain worthy the name in the first crop. Again in agreement with the results of the other series, considerable discrepancy was found in duplicate pots so far as yields are concerned. In the case of the root yields, we have also large discrepancies between duplicate pots. However this may be, the straw yields in the treated pots of the first crop surpass those of the untreated pots in almost all cases, even if the higher figure for straw yields of the control pots be employed as a criterion. This is not so for the yields of roots; and while average yields show definite stimulation by  $\text{FeSO}_4$  for root production of barley plants on the greenhouse soil, single values from duplicate pots do not justify any conclusions of that nature. Despite all this, there is certainly no reliable evidence of definite toxic effects on the part of  $\text{FeSO}_4$  to barley plants under the conditions of this experiment. In general, therefore, the results of the  $\text{FeSO}_4$  series are not unlike those of the  $\text{CuSO}_4$  series, and the  $\text{ZnSO}_4$  series on the greenhouse soil so far, at least, as the first crop is concerned.

#### SECOND CROP

It will be remembered again that the amounts of  $\text{FeSO}_4$  employed for the first crop were doubled before planting the second crop. On studying the yields of the latter, one is at once struck by the strong parallelism in effect exerted on the barley plants by  $\text{ZnSO}_4$  and  $\text{FeSO}_4$  in the second crop. Both stimulate total dry-matter production in the very low concentrations and yet the discrepancies between the actual amounts of salts used in the two cases are of course very large. Besides, both seem to stimulate root development and, very slightly, the production of straw, at certain concentrations. Again, there appears to be indirect evidence that most of the salt applied is not only rendered insoluble, as is probably the case with  $\text{FeSO}_4$ ,

but that much of the salt remaining is adsorbed by the soil and becomes inactive so far as the barley roots are concerned.

### THIRD CROP

In table VIc, which sets forth the yields of dry matter obtained in the third crop of the  $\text{FeSO}_4$  series, we find some data of unusual interest. Despite discrepancies in the weights of dry matter obtained in duplicate pots, there can be little question that the higher concentrations of  $\text{FeSO}_4$ , beginning with 1.4 per cent, definitely stimulate straw production, while the lower concentrations employed, less definitely but probably without doubt, depress it. Grain production, on the other hand, seems to have been stimulated in the third crop by all concentrations of  $\text{FeSO}_4$  employed, and the yields are high enough and agree well enough in the duplicates to justify that conclusion. In the case of the roots, still another effect was probably induced by  $\text{FeSO}_4$ . No stimulation can be definitely noted, yet the toxic effect, if any, is small and apparent in very few instances. In the case of the total dry matter produced, marked stimulation seems to have been obtained at concentrations of  $\text{FeSO}_4$  respectively of 1.4 per cent, 1.6 per cent, and 2 per cent. When compared with the third crop of the  $\text{CuSO}_4$  and  $\text{ZnSO}_4$  series, the third crop of the  $\text{FeSO}_4$  series stands out sharply. It gives stimulation only at the higher concentrations, the  $\text{ZnSO}_4$  gives no stimulation and almost positive toxicity throughout, and the  $\text{CuSO}_4$  gives stimulation almost throughout the whole series in the third crop. While all three of the salts may be quite harmless and even stimulating in relatively small quantities, they manifest very definite and specific characters when employed in higher concentrations and when results are obtained on the same soil for more than one season.

### LEAD SULFATE—GREENHOUSE SOIL

Entirely unlike the three salts thus far discussed,  $\text{PbSO}_4$  exercises what appears to us to be a definitely toxic effect throughout the first crop. This observation must be considered separately for every crop. It should be noted that unlike the

copper, zinc, and iron salts,  $\text{PbSO}_4$  was applied once only prior to planting the first crop.

#### FIRST CROP

Like  $\text{ZnSO}_4$  and  $\text{FeSO}_4$ , the  $\text{PbSO}_4$  was tested in the greenhouse soil only. The yields obtained in the  $\text{PbSO}_4$  series are given in table VIIa, VIIb, and VIIc. They will be discussed in conjunction with the comment already made with reference to the aspect of the plants in the  $\text{PbSO}_4$  series. It will be noted there that the plants possessed little rigidity, were deep green in color, and in general assumed a sprawling or prostrate, instead of an erect, habit of growth. This was a result of some specific reaction of  $\text{PbSO}_4$  and was exerted even though only small quantities of the salt could have existed in the soil solution, owing to the insolubility of the salt. It should also be observed in this series, as it has been in the others, that the quantity of  $\text{PbSO}_4$  employed seemed to have little relation to its toxic effects on the yields of straw. The lack of grain production has already been explained in other discussions above and is connected not with any salt treatment, but with the condition of the greenhouse soil itself, of which more detailed discussion has been given.

Root production was particularly affected in a deleterious manner by  $\text{PbSO}_4$  in the first crop. Roughly speaking, it was reduced in the  $\text{PbSO}_4$  treated pots by more than 60 per cent of the yield obtained on the untreated or control pots. In other words, in this series, as in many others, root production and straw production run almost parallel. This is further evidenced by the uniformly depressing effect of  $\text{PbSO}_4$  regardless of the quantity in which it was employed. We find therefore in  $\text{PbSO}_4$  (and in Pb, because all the sulfates used have a common anion) a substance which in the first crop on the greenhouse soil exhibits characteristics totally different from those of copper, zinc, and iron under similar circumstances. Thus while the three salts last named show definite powers of stimulating barley growth in the first crop on the greenhouse soil, lead very markedly depresses the growth of that plant under the same conditions.

## SECOND CROP

In the second crop quite different conditions obtain with respect to the effects of  $\text{PbSO}_4$ . While nearly all of the higher concentrations of the series are still toxic to barley, three of the lower concentrations, including 600 p. p. m., are distinctly stimulating to that plant. If it were not for injury to the plants by mice, the concentrations of 200 p. p. m. and 400 p. p. m.  $\text{PbSO}_4$  would doubtless have shown as much stimulation as the others just mentioned. In other words, taking the total dry matter produced, it seems true beyond cavil that in the second crop on the greenhouse soil,  $\text{PbSO}_4$  in very considerable concentrations acts as a stimulant to barley growth.

With reference to the separate fractions of the total dry matter produced in the second crop of the  $\text{PbSO}_4$  series, we note some interesting facts. In the first place, no grain was produced in the second crop of the lead series. This is very difficult to explain, since the control pots and the treated ones behaved similarly in that regard. In view of our statements in the introductory portion of this paper, we can scarcely believe that the mere location of the plants of this series in a somewhat shaded part of the greenhouse can account for the discrepancy. The root yields were nearly all depressed by the action of  $\text{PbSO}_4$  in the second crop. The exceptions to this rule were in isolated pots with no duplicates to confirm them. It would therefore seem that  $\text{PbSO}_4$  is toxic to the root development of the barley plant even in the second crop, in spite of its stimulating effect on the straw yield at certain concentrations. Such effect of  $\text{PbSO}_4$  is unlike that of any of the other salts, which, at least at a number of concentrations, stimulate root development, particularly so in the case of  $\text{CuSO}_4$  and  $\text{ZnSO}_4$ .

## THIRD CROP

A progressive improvement may be seen in the soil treated with  $\text{PbSO}_4$  as crop follows crop. Just as the second crop gave very much better results than the first, so the third crop gave very much better results than the second. In the third crop the stimulation to the growth of barley exerted by  $\text{PbSO}_4$  is, however, the most striking, since it obtains particularly at

the higher concentrations of  $\text{PbSO}_4$ . As was the case with some of the other salts in other crops,  $\text{PbSO}_4$  seems to be toxic in the third crop at the low concentrations at which it stimulated growth in the second crop. On the other hand, it stimulates growth as above stated in the third crop at some of the higher concentrations at which it was toxic in the second crop.

However, most of the stimulating influence of  $\text{PbSO}_4$ , and perhaps all of it, in the third crop affected the straw production and not the root yields. This is again at variance with the results obtained in many of the other series above described, in which the usual condition was a parallelism between the effects exerted by a salt on the different fractions of the total dry-matter yields. Thus very good straw yields were obtained in most of the pots of the series in the third crop and instances of increases over those of the control pots were numerous, but no definite evidence of such stimulation in the case of the roots could be noted. In the case of the grain, on the other hand, the higher concentrations of  $\text{PbSO}_4$  seemed to be as definitely stimulating as they did in the case of the straw yields. This is in almost entire harmony with  $\text{FeSO}_4$  in the third crop, but has little resemblance to the corresponding  $\text{CuSO}_4$  and  $\text{ZnSO}_4$  series.

#### POTASH ALUM—GREENHOUSE SOIL

This salt was tested in these experiments because it had been proposed that if it was not detrimental to soils and crops, it could be employed as a source of potash for fertilizers. It could be cheaply obtained in all probability by treating granitic rock containing adequate percentages of potash, with  $\text{H}_2\text{SO}_4$ , which can be manufactured in large quantity by the important smelter plants through the oxidation of  $\text{SO}_2$  fumes. In view of the foregoing, potash alum was applied, as indicated in tables VIIIa, VIIIb, and VIIIc, which are given below, on the basis of a certain number of pounds per acre, beginning with 300 pounds  $\text{K}_2\text{O}$  per acre in the form of  $\text{KAl}(\text{SO}_4)_2 \cdot 12\text{H}_2\text{O}$  and going up to 2000 pounds  $\text{K}_2\text{O}$  per acre in the same form.

## FIRST CROP

Table VIIIa shows clearly that the application of potash alum in the first crop was distinctly stimulating to the barley plant so far as the production of total dry matter is concerned. The degree of stimulation is not unlike that of  $\text{CuSO}_4$ ,  $\text{ZnSO}_4$ , and  $\text{FeSO}_4$  in the first crop and, again, seems to be about the same with the lower as with the higher concentrations. When we consider the root yields separately from the straw yields we find, however, that the former were not increased by the potash alum treatment, though they were scarcely depressed with any concentration of the salt.

## SECOND CROP

With the concentration of potash alum doubled in the second crop, the marked evidences of its stimulating effect on barley growth are still manifestly present. The entire series of treated pots, when averages of total dry matter produced are taken as the criterion, gives results far superior to those of the control pots, even though there is variation among the latter and among the treated pots in duplicate cultures. So far as the production of the total dry matter of barley is concerned, there appears to be no evidence in the second crop and very little, if any, in the first, of any toxic properties of potash alum.

We may now consider briefly the separate parts of the total dry matter as affected by the potash alum. The yield of straw is without exception greater in the treated than in the untreated pots of the second crop of the potash alum series. That part of the total dry matter has therefore been very materially increased by the potash alum application. The grain yields in the absolute were of very great magnitude and amounted in many cases to as much by weight as the dry matter of the straw. In some cases they even excelled the latter. This is analogous to the condition of the second barley crop in the  $\text{CuSO}_4$  series, which was the only one of the other series manifesting as high a grain production. Not only, however, were the grain yields large in the absolute, but they indicated clearly the stimulating effect of potash alum on their production, since all the treated pots yielded much more grain than the control pots.

In the second crop, on the other hand, as in the first, the larger amounts of potash alum were neither inferior nor superior to the smaller amounts in increased production of straw and grain, but were of about the same influence throughout. Consistent with the effect of potash alum on the straw and grain yields was that on the root yields. The latter were, throughout the whole series in the second crop, increased by the potash alum applications and, as in the cases of straw and grain, independently of the amounts of potash alum employed. We have, therefore, another phase of analogy between the potash alum and the  $\text{CuSO}_4$  series in the second crop which seems only to make the resemblance stand out in greater relief. The production of every part of the plant in the second crop was stimulated by both potash alum and by  $\text{CuSO}_4$ , but not by the other sulfates employed.

#### THIRD CROP

Wholly at variance with the effects just noted are those observed in the third crop of the potash alum series. So far from stimulating the growth of barley in all respects, as it did in the first and second crops, and particularly in the latter, potash alum in any and all concentrations depresses the growth of barley when the yields of total dry matter are used as a basis of comparison. This is true also for the straw and root yields taken separately, with the possible exception of the straw yield with the lowest concentration of potash alum. In the case of the grain yields, however, no indubitable evidence of a depressing effect by the potash alum is at hand. It is indeed not impossible that definite though small effects of potash alum stimulating to grain production in the third crop might be allowed in some of the concentrations of the salt employed. The explanation of this striking change in the effects of potash alum in two successive crops is obviously not simple, though several possible explanations immediately suggest themselves. It is probable that the most favorable explanation would be over-stimulation of plant growth in the first two crops and the removal of most of the easily available bases in the soil, leaving an impoverished soil condition and perhaps a so-called "physiological acidity,"



which would of course react deleteriously on the development of the barley plant. Again, the washing out of the salt by irrigation may have caused physical conditions in the soil which are inimical to the proper air and water supply for both plants and the soil bacteria. The first conception is the one employed to a considerable extent by the "old-line soil chemist" to explain the depressing effects on soil fertility of the large and continued use of gypsum. The second is a condition demonstrated in this laboratory recently<sup>7</sup> to be of considerable importance. Further discussion will be accorded this subject in a general comparison given below of our results with those of others. In general it may be added that the results of the third crop in the potash alum series are more in keeping with those of the  $\text{ZnSO}_4$  series than with those of any other series discussed.

#### MANGANESE SULFATE

After our work on the effects of the compounds mentioned on barley plants had been under way for one season, it was deemed advisable to inaugurate some new experiments, using manganese salts. The latter it will be remembered were represented by  $\text{MnSO}_4$  in preliminary experiments by F. H. Wilson and the senior author, which are cited above. Owing to the fact that the preliminary experiments with manganese had shown the latter to be comparatively innocuous, and even stimulating at considerable concentrations, for barley and vetch, larger amounts of manganese than of copper and zinc were employed. Both  $\text{MnSO}_4$  and  $\text{MnCl}_2$  were tested. Each of these salts will be considered separately, and tables IXa, IXb, and IXc, which follow, give the plan and the results of the experiments with  $\text{MnSO}_4$ . In the case of both manganese salts, only one application was made, and that was prior to the first crop.

#### FIRST CROP

It becomes at once clear from an examination of table IXa that we can find in the first crop no indubitable evidence of

<sup>7</sup> Univ. Calif. Publ. Agri. Sci., vol. 1, no. 10, p. 291.

toxicity for barley of  $\text{MnSO}_4$  even when amounts of that salt equivalent to 0.6 per cent of the dry weight of the soil were used. On the other hand, the stimulating effect of  $\text{MnSO}_4$  for barley in the first crop on the greenhouse soil appears to be clearly evident. This is particularly true for the first three concentrations, amounting respectively to 500, 1000, and 1500 p. p. m. At concentrations exceeding 1500 p. p. m.  $\text{MnSO}_4$ , the stimulation is only slight, and three concentrations—namely, 3500 p. p. m., 4000 p. p. m., and 5500 p. p. m.  $\text{MnSO}_4$ —possibly depress barley growth to some extent. The latter effect can scarcely be taken as indicating definite toxicity, however, since, as above pointed out, even the highest concentration employed (6000 p. p. m.  $\text{MnSO}_4$ ) appeared to stimulate barley growth slightly, and the toxic evidences referred to are noted at concentrations which lie between slightly stimulating concentrations on both sides. At any rate, we have no evidence of the toxicity of  $\text{MnSO}_4$  in the first crop until concentrations equivalent to 3500 p. p. m. of  $\text{MnSO}_4$  are reached.

When the root and straw yields are considered separately in the first crop, some interesting observations may be made which are not possible when the dry matter is considered as a whole. For example, stimulation to root development in the first crop of the  $\text{MnSO}_4$  series is apparent only in the first three concentrations above noted as giving the largest yields of dry matter. Moreover, the straw yields are also distinctly higher at those same concentrations. But whereas  $\text{MnSO}_4$  gives evidence of toxicity to root development, either slightly or definitely, at all concentrations tried above 1500 p. p. m., it continues beyond that concentration to be slightly stimulating to straw production.

In comparison with the other salts above described,  $\text{MnSO}_4$  is distinctly superior in the magnitude of its stimulating effects. The only other salt which manifests some resemblance to  $\text{MnSO}_4$  in that respect is  $\text{CuSO}_4$ . Since the concentrations of these two salts here employed, however, are very different from each other in the two cases, no more detailed comparison would be wholly justified.

## SECOND CROP

When the total dry matter of the second crop in the  $\text{MnSO}_4$  series is considered (table IXb), we find that not only has the stimulation noted in the first crop disappeared, but that an actually definite toxicity has supplanted it. Moreover, such toxicity is as marked with the lowest as it is with the highest concentrations of  $\text{MnSO}_4$ , and it is even possible that the former definitely surpass the latter in that respect. Again, as in some preceding cases with other salts, the total dry-matter yields do not give a complete picture of the effects of  $\text{MnSO}_4$  on barley growth. Thus if we consider the straw, grain, and root yields separately, we find data leading to conclusions slightly different from those above. For example, whereas both the grain and root production are definitely depressed at all concentrations of  $\text{MnSO}_4$  in the second crop, this is not so for the straw yields. The latter are in many instances, including the cultures of the highest concentrations of  $\text{MnSO}_4$ , increased by the effects of the salt. Were it not for the lack of agreement in some of the duplicates, we might add more emphatically that straw yields are markedly stimulated by  $\text{MnSO}_4$  in the second as in the first crop on the greenhouse soil. This seems particularly true at the higher concentrations of the latter salt, but is also apparent at some lower concentrations. Since, therefore, no grain was produced in the first crop, and since only three of the lowest concentrations of  $\text{MnSO}_4$  in it gave stimulation to root development, it seems not unreasonable to consider that the results of the second crop in the  $\text{MnSO}_4$  series are, in the large, not essentially different from those of the first crop. The outstanding result is the stimulation to straw production which is noted, and that is different in degree only, not in kind, in the two crops here considered. Despite all this, however, we do not attempt to disregard the differences which characterize the effects of  $\text{MnSO}_4$  in the first and second crops as above pointed out, but we regard them as of minor significance.

When we compare  $\text{MnSO}_4$  in the second crop with other salts under similar circumstances in the greenhouse soil, we find that it has but little in common with them. It approaches perhaps most closely the behavior of  $\text{PbSO}_4$  in the second crop, but is

different from it in several important particulars. As a general thing, the other salts still give more stimulating effects in the second crop so far as the total dry-matter production is concerned, but this is not true in any instance of the second crop of the  $\text{MnSO}_4$  series. It should be borne in mind, however, that the manganese series is not comparable with the others except possibly with the lead series, because only one treatment, prior to the first crop, was given.

#### THIRD CROP

The depressing effect exerted by  $\text{MnSO}_4$  in the second crop of barley, at least so far as the grain and straw yields are concerned, appears to have been merely an ephemeral one. There was not only a total disappearance thereof in the third crop, but an actually stimulating effect seems to have replaced it; and to have extended to straw, grain, and root production and was not confined, as in the second crop, merely to straw production in part of the series. Moreover, the stimulating effect of the  $\text{MnSO}_4$  appears to have extended throughout all concentrations and would seem to have been greatest at the medium high concentrations, as is indicated in table IXc. While much better agreement between duplicate determinations could have been desired, the clear superiority in yield of the majority of treated pots, when compared with the controls, leaves scarcely any room for doubt that we are here confronted with real cases of stimulating effects. The results are the more interesting and striking since large concentrations of  $\text{MnSO}_4$  are involved. The results call for further observations on the apparent reversal of results between the second and third crops and between the second and first crops. Unfortunately, no definite leads are in our possession which would aid us in answering this question. Theoretically, however, it would seem possible to explain the facts as follows: In the first crop the large quantity of organic matter present in the soil brings about the adsorption of the  $\text{MnSO}_4$ , and leaves the active soil solution relatively dilute in that salt. This low concentration acts as a stimulant to both the higher plants and the soil flora and induces an increased yield. After one season of exposure to sun and cultivation, the soil loses a

considerable portion of its organic-matter supply and therefore possesses a much smaller surface for adsorption of  $\text{MnSO}_4$ . Hence the usable portion of the soil solution would tend to become more concentrated with respect to that salt and induce depressions in yield of roots and grain. By the time the third crop is planted, thorough oxidation of the  $\text{MnSO}_4$  has occurred and most of the manganese is rendered insoluble, thus leaving again only a small quantity of the salt in the soil solution. This acts as a stimulant, as it did in the first crop, and induces an increased yield again. This explanation, while open to question in one or two important respects, may prove of some assistance in the ultimate clearing up of the somewhat perplexing facts which are here considered. Other explanations, involving the relationship of  $\text{MnSO}_4$  to the soil colloids and to other phases of the soil solution besides that above mentioned, offer themselves at this time, but they must all await the further study of fundamental principles of plant physiology before they can be considered to advantage. Irrespective of the theoretical arguments which may account for the results obtained in the  $\text{MnSO}_4$  series, the striking facts relating to the changes in effect on three successive crops of a given salt application made prior to the first planting are of great practical moment. Not only do they render of doubtful value for practical purposes one season's results on the effects of salts on crops, but they cause one to wonder if anything less than five successive crops should ever constitute sufficient evidence upon which to base a judgment. Taking all of our results together, it may be said in general that  $\text{MnSO}_4$  is to be regarded, for a limited period at least, as definitely stimulating to barley growth in soils.

#### MANGANESE CHLORIDE

As pointed out above,  $\text{MnCl}_2$  was tested along with  $\text{MnSO}_4$ , so that manganese in different compounds might be studied for itself as well as in comparison with other elements. The experiment was arranged similarly to that of the  $\text{MnSO}_4$  series, and details with respect to it, together with the results obtained, are set forth in tables *Xa*, *Xb*, and *Xc* for the three crops grown.

## FIRST CROP

Table Xa shows the striking effects of concentrations of  $\text{MnCl}_2$  on barley growth in the greenhouse soil during the first crop. Whereas the first three concentrations of that salt, namely 500 p. p. m., 1000 p. p. m., and 1500 p. p. m., give very marked stimulation to barley growth (far more indeed than that given by similar concentrations of  $\text{MnSO}_4$ ), amounts in excess of 1500 p. p. m.  $\text{MnCl}_2$  are very markedly toxic. This toxicity increases strikingly with the increase in concentration in  $\text{MnCl}_2$  beyond 2500 p. p. m., until at a concentration of 6000 p. p. m. almost no growth is obtained. Even the difference between 1500 p. p. m. and 2000 p. p. m. in the soil means a change from a high degree of stimulation for barley production to a marked toxicity and a decrease of about 50 per cent in the yield. No series of salt concentrations studied by us and reviewed above gave anything like the sharpness of manifestation of toxicity that is noted in the first crop of the  $\text{MnCl}_2$  series. We are evidently dealing again with the acute toxicity of chlorine for living cells which we have on other occasions pointed out in various connections. This is true, if we may repeat again, despite the fact that at the lower concentrations, chlorine may, as is strikingly exemplified in table Xa, give astounding evidences of stimulation to barley which surpasses any noted above with other and more uniformly stimulating substances.

When we study straw and root yields separately we find that, in general, the effects of  $\text{MnCl}_2$  are similar with respect to both in the first crop. The roots are, to be sure, only slightly stimulated in growth in the first three concentrations employed, whereas the tops are enormously stimulated. When, however, the toxicity of  $\text{MnCl}_2$  becomes apparent, it is equally striking in the roots and tops, as the figures in the table clearly show. While in some respects, therefore, and particularly as regards stimulation, the  $\text{MnCl}_2$  behaves like the  $\text{MnSO}_4$  in the first crop and the first three concentrations, it is totally different from the latter salt in giving marked evidences of toxicity at concentrations in excess of 1500 p. p. m. Nevertheless  $\text{MnSO}_4$  still continues to stimulate growth, even though it does so very slightly throughout the series. On the other hand, although the resem-

blance between the behavior of  $\text{MnSO}_4$  and  $\text{MnCl}_2$  in the first crop is limited in extent,  $\text{MnCl}_2$  resembles  $\text{MnSO}_4$  much more in its effects on barley growth in the first crop than it does any of the other salts under similar circumstances. Again, we are obliged to stop with this general comparison owing to the high concentrations of  $\text{MnCl}_2$  employed, as compared with the relatively much lower or much higher concentrations of the other salts employed.

#### SECOND CROP

When the total dry matter is considered (see table Xb), the second crop of the  $\text{MnCl}_2$  series gives the latter salt a reversal of form. At the first three concentrations at which it notably stimulated the production of dry matter in the first crop, it becomes decidedly toxic in the second crop. On the other hand, at the concentrations above 1500 p. p. m., at which it was acutely toxic in the first crop,  $\text{MnCl}_2$  is stimulating when the total yields of the treated as against those of the untreated pots are considered. Such marked reversal of effects of  $\text{MnCl}_2$  between two succeeding crops on the same soil needs further attention under the general discussion below. Following the procedure employed in the case of the other series, we may now study separately the yields of straw, grain, and roots as given in table Xb. Taking the straw yields first, we find that they were, in all cases but one or possibly two in the series, much larger than those of the control pots, and that at concentrations in excess of 1500 p. p. m. the average yield of straw was nearly twice as great as that of the control pots. While different in degree, therefore, this effect of  $\text{MnCl}_2$  in the second crop is very similar in kind to that exerted by  $\text{MnSO}_4$  in the second crop with respect to the yield of straw.

In the case of the grain, however, we find totally different conditions, for here only one case of stimulation is noted and that, owing to the great discrepancy in the duplicates, is an unsafe one to accept. In excess of 3000 p. p. m.,  $\text{MnCl}_2$  manifests a very marked toxicity so far as grain production is concerned, until at 6000 p. p. m. very little or no grain is produced. This result is again different only in degree, not in

kind, from that of the corresponding one in the  $\text{MnSO}_4$  series. Respecting root yields, the toxic effects of  $\text{MnCl}_2$  in the second crop are apparent throughout the whole series. While the decreases are not quite so great at the lower concentrations of  $\text{MnCl}_2$  as they are at the higher concentrations, they are not far different, and in general amount to from 40 to 60 per cent of the amount yielded by the control pots. We see in the root yields, therefore, a further analogy between the second crop of the  $\text{MnCl}_2$  series and that of the  $\text{MnSO}_4$  series. In brief, it should be observed that while wide discrepancies in total yields of dry matter are noted between the second crops of the  $\text{MnCl}_2$  and  $\text{MnSO}_4$  series, the discrepancies are superseded by striking resemblances when the straw, grain, and root yields are compared separately in the two series. Since the differences seem to be those of degree only, is it not possible that we have here the dominant manifestations of the effects of manganese, which are only slightly modified by the element or elements combined therewith? If this were not the case, would we not expect to find much larger discrepancies between the two series in question, based on the specifically different effects of the  $-\text{Cl}$  and the  $-\text{SO}_4$  ions on barley growth?

#### THIRD CROP

The stimulating powers of manganese, as exemplified in the effects of  $\text{MnSO}_4$  ions on the third crop of barley, are again manifest but very much more strikingly in the third crop of the  $\text{MnCl}_2$  series. While the yields of duplicate pots still fail to agree closely in a number of the salt concentrations tested, they show a much better agreement than those of the  $\text{MnSO}_4$  series. At any rate, there can be no doubt of the stimulating effects of manganese chloride for barley grown in the greenhouse soil even in the third crop. Again, as was the case in the  $\text{MnSO}_4$  series, the  $\text{MnCl}_2$  stimulates the production of all parts of the plant and not merely of any one portion of the dry matter thereof. Thus, for example, whereas there was practically no stimulation to grain production in the second crop of the  $\text{MnCl}_2$  series, the third crop shows such stimulation markedly throughout the series. In no case, further, so far as the total



dry weight produced is concerned, did any of the treated pots produce so low a yield as the control pots, when averages are considered. The great immunity to chlorine which the plants in the third crop of this series manifest is very difficult to explain. In general, however, the changes in the effects of  $\text{MnCl}_2$  from one crop to another are much the same in nature as those of the  $\text{MnSO}_4$  series which have been discussed more in detail above. It looks obvious that we are dealing primarily in both manganese series with the effects of the kation rather than with those of the anions, though, to be sure, specific effects of the latter do not seem to be wanting. The balance of the data presented in table Xc speaks for itself.

## COMPARISON OF OUR RESULTS WITH THOSE OF PREVIOUS INVESTIGATORS

It is quite unnecessary to review in detail the results of the numerous investigations which bear on the subject in hand, particularly those relating to copper and its influence on living organisms. Although, therefore, we are herewith citing a very extensive bibliography, we shall make no attempt at reviewing all of the investigations which have been carried out. It does seem desirable, however, to compare in general the results of our investigations with those of other researches in the hope that we may thereby arrive at some definite understanding, now that so much experimental work has been accomplished, as to the real status of the salts in question in the realm of plant physiology. In order to simplify such discussion, we shall take up the different so-called toxic metals separately.

### COPPER SULFATE

As pointed out above, the bibliography on the subject of copper and its effects on plants is very extensive. One needs but to turn to the complete reviews of it by Czapek,<sup>8</sup> Pfeffer,<sup>9</sup>

<sup>8</sup> *Biochemie der Pflanzen*, vol. 2, p. 910, Jena, 1905.

<sup>9</sup> *Pflanzenphysiologie*, vols. 1 and 2, Leipzig, 1897 and 1901.

and Brenchley<sup>10</sup> to be confirmed in that opinion. The general impression given by the reviewers is that so far as plants are concerned, copper is to be regarded as a distinctly toxic substance. To quote Brenchley from the work above cited, for example: "Altogether, after looking at the question from many points of view, one is forced to the conclusion that under most typical circumstances, copper compounds act as poisons to the higher plants, and that it is only under particular and peculiar conditions and in very great dilutions that any stimulative action on their part can be clearly demonstrated." This statement is not qualified with respect to the kind of medium employed for testing the effects of copper on plants. But whether it be applied to solution or to soil cultures, it would scarcely seem to be adequately supported by experimental evidence, and particularly is this true regarding soil cultures. In solution cultures, copper in various compounds was found to be toxic to the higher plants by Otto,<sup>11</sup> Haselhoff,<sup>12</sup> Coupin,<sup>13</sup> Kanda,<sup>14</sup> True and Gies,<sup>15</sup> True and Oglevee,<sup>16</sup> Jensen,<sup>17</sup> Brenchley,<sup>18</sup> Heald,<sup>19</sup> Harter,<sup>20</sup> and Haywood.<sup>21</sup> While exceedingly high dilutions of copper salts were employed by some of these investigators, the possibility still exists in their work that the merest traces of copper may have acted as stimulants. Moreover, in the case of Jensen's work the evidence on the toxicity of very dilute solutions of copper salts is really negative, since he emphasizes principally the fact that no stimulation was observed with  $\text{CuSO}_4$  in solution cultures.

<sup>10</sup> *Inorganic plant poisons and stimulants*, Cambridge, 1914.

<sup>11</sup> *Ztschr. Pflanzenkrankh.*, vol. 3, no. 6; *Bot. Cent.*, 56, p. 340; *E. S. R.*, 5, p. 649.

<sup>12</sup> *Landw. Jahrb.*, 21, p. 263; *E. S. R.*, 3, p. 499.

<sup>13</sup> *Comptes Rendus Acad. Sci.*, Paris, 127, p. 400; *E. S. R.*, 10, p. 611.

<sup>14</sup> *Jour. Col. Sci. Imp. Univ. Tokyo*, 19, p. 47; *Bot. Cent.* 95, p. 538; *E. S. R.*, 16, p. 228.

<sup>15</sup> *Bull. Torr. Bot. Club*, 30, p. 390.

<sup>16</sup> *Bot. Gaz.*, 39, p. 1; *Science*, 19, p. 421.

<sup>17</sup> *Bot. Gaz.*, 43, p. 11.

<sup>18</sup> *Inorganic plant poisons and stimulants*, Cambridge, 1914.

<sup>19</sup> *Bot. Gaz.*, 22, p. 125.

<sup>20</sup> *U. S. Dept. Agr., Bur. Pl. Ind., Bull.* 79, p. 40.

<sup>21</sup> *U. S. Dept. Agr., Bur. Chem., Bulls.* nos. 89, 113, and 113, revised.

Opposed to the findings of the investigators just named were those which showed evidence of stimulating effects of copper salts to plants in solution cultures. Among these investigators were Tschirch,<sup>22</sup> Montemartini,<sup>23</sup> and Forbes.<sup>24</sup> So far as germination of seeds is concerned, Effront<sup>25</sup> also noted the stimulating effect of copper. Owing to conflicts in the results obtained by different investigators working with copper in solution cultures, one seems scarcely justified in subscribing to the statement above quoted from Brenchley, even if it were made to apply only to solution cultures. As Dr. Brenchley herself admits, there is no absolutely satisfactory method for determining whether or not a certain substance is toxic or stimulating to plants. But from the theoretical standpoint of ascertaining how the protoplasm of the plant is affected by a given substance, if at all, the solution-culture method is the only one involved, since the other methods are confessedly not intended to show anything more than effects of substances on plants under conditions closely approximating the natural. If, then, the solution culture method is the only one among those at present known that is suitable for studying the effects of different chemicals on plant growth in a more or less intimate way, why do we obtain the conflicting results above noted with respect to the effects of copper on plants? The answer to this question is to be found in a number of circumstances surrounding the manipulation of the solution-culture method. Some investigators use distilled water, others use tap water, still others physiologically balanced solutions of a large variety. For reasons well known to plant physiologists, the results of such different media among the solution cultures must show wide discrepancies. If, however, the claim is made that all media but pure distilled water be discarded in such work, owing to the factors of salt antagonisms which enter into salt solutions to vitiate results, a very strong counter-claim can be made. The protoplasm of plant cells is not in a natural medium when it is placed in distilled water, and

<sup>22</sup> Abstract in Chem. Ztg., 18, p. 320; E. S. R., 6, p. 872.

<sup>23</sup> Staz. Sper. Agr. Ital., 44, p. 564.

<sup>24</sup> Results soon to be published, Univ. Calif. Publ. Agri. Sci.

<sup>25</sup> Compt. Rend. Acad. Sci. (Paris), 141, p. 626; E. S. R., 18, p. 126.

hence it may manifest distress and weakness which under natural conditions might be quite impossible. Owing to osmotic influences, the plant would lose salts and other substances to the distilled water more quickly and in larger quantity than to tap water or to a balanced solution. It would therefore be more subject to weakening or to the absorption of toxic materials in the former than in the latter medium. In other words, under such circumstances copper, for example, would merely exaggerate the untoward conditions for plant growth, while it might have no power to affect the plant under more favorable conditions. Again, seeds are not usually allowed to germinate in the solution which is to be tested in the cultures, but in a medium of a harmless nature. Does not sudden removal to salt-solution cultures render them less immune to certain substances than if they had been allowed to accustom themselves from the beginning to a given salt?

We do not desire to give the impression from these arguments that we deprecate the use of the solution-culture method. On the contrary, we think it of great value in the study of many fundamental problems and also for obtaining relative data. When, however, one attempts to use it in drawing absolute conclusions for purposes of application to such a subject as that under consideration, it falls as far short of throwing light on the actual effects of a given substance on plant protoplasm (as the latter is situated under natural conditions), as does any other method of study now employed. We believe that the conflicts in the results just reviewed are perhaps explicable on one of the bases above discussed; and since no modification of the solution-culture method is free from serious objection, we must accord equal value to all results of reliable investigators. Consequently we arrive at the conclusion that in the experiments above cited there is no absolute evidence that copper is or is not stimulating to plant protoplasm in solution cultures. While there appears to be more evidence that copper is toxic under the conditions and in the concentrations named than that it is stimulating, we cannot admit that the plant has been tested in any two of the experiments under essentially the conditions of its natural

habitat. Since plants are, after all, to be found growing naturally only in soils, it cannot be a matter of indifference to us, in attempting the study of the effect of a certain substance or substances on them, whether they are supplied with normal conditions for their development or not.

Proceeding now to an examination of the results obtained by other investigators on the effects of copper on plants grown in soil or sand instead of solutions we find many interesting observations. Injurious effects of  $\text{CuSO}_4$  at the rate of about 400, 800, and 1600 pounds per acre to potatoes and beans were noted by Steglich,<sup>26</sup> but he failed to observe such toxic effects on the same soil to strawberries or fruit trees. Haselhoff<sup>27</sup> claims also to have noted injury to grass, beans, and other plants from smelter smoke containing copper. Owing to other conflicting factors concerned in smelter-smoke injury, Haselhoff's results are open to serious criticism. Simon<sup>28</sup> experimented with oats and mustard on garden soil, clay, and sand, and used amounts of  $\text{CuSO}_4$  varying from 0.01 per cent to 10 per cent. His statements imply that copper was toxic throughout, with the oat plants showing more resistance than the mustard, and that  $\text{CuSO}_4$  was least toxic in garden soil and most toxic in the sand. Opposed to the three cases just cited are numerous results showing the stimulating effects of copper to plant growth in soils. We find among these the results obtained by Girard,<sup>29</sup> Kanda,<sup>30</sup> Jensen,<sup>31</sup> Voelcker,<sup>32</sup> Forbes,<sup>33</sup> and Sachser.<sup>34</sup> A large number of observations have also been made on the stimulating effects, or lack of any effect, of copper sprays, and in other ways of the effect following direct contact of the copper solution with plant cells, among which may be mentioned those of Frank and

<sup>26</sup> Ber. Tat. Landw. Abt. K. Vers. Stat. Pflanzenkult, Dresden, p. 4, 1903; E. S. R., 16, p. 133.

<sup>27</sup> Fühling's Landw. Ztg., vol. 57, no. 18, p. 609; E. S. R., 20, p. 831.

<sup>28</sup> Landw. Vers. Stat., 71, p. 417; E. S. R., 22, p. 439.

<sup>29</sup> C. R. Acad. Sci., Paris, 120, p. 1147; E. S. R., 7, p. 99.

<sup>30</sup> Jour. Col. Sci. Tokyo Imp. Univ., 19, p. 47; E. S. R., 16, p. 228.

<sup>31</sup> Bot. Gaz., 43, p. 11; E. S. R., 18, p. 625.

<sup>32</sup> Jour. Roy. Agr. Society, England, vols. 73, 74, and 75, Report for 1912, 1913, and 1914.

<sup>33</sup> Univ. Calif. Publ. Agr. Sci., 1, no. 12, 1917.

<sup>34</sup> Cent. Agr. Chem., 33, p. 533; E. S. R., 16, p. 865.

Kruger,<sup>35</sup> MacDougal,<sup>36</sup> Chuard and Porchet,<sup>37</sup> Demoussy,<sup>38</sup> Prandi,<sup>39</sup> Olive,<sup>40</sup> and Molinari and Ligot.<sup>41</sup> In addition to all these results, which show either no toxicity or decidedly stimulating effects on plants from the use of copper (usually  $\text{CuSO}_4$ ) in soil, there are extant a number which testify to the high resistance of plants in soil to extremely large amounts of copper (Such, for example, as from 2 per cent to 5 per cent of the dry weight of the soil). Among these may be mentioned the observations of Van Slyke<sup>42</sup> and Pammel.<sup>43</sup>

All of these findings render it extremely improbable that copper in soil, can at any time be considered definitely toxic in relatively small quantities (say, below 0.10 per cent of the dry weight of the soil). On the contrary, the evidence seems very well established that positive stimulation of plants may be induced through the use of small quantities of copper (say, from 0.01 to 0.05 per cent of the dry weight of the soil), in the form of  $\text{CuSO}_4$  particularly, and possibly also in other forms. Our investigations as discussed would seem to confirm and be confirmed by earlier investigations of the senior author and F. H. Wilson and by numerous other experiments carried out in different parts of the American and European continents and in England. These observations would appear therefore to refute the conclusion of Dr. Brechley which is above quoted and to point clearly, through the added data which we have submitted, to the conclusion that copper in the form of  $\text{CuSO}_4$  is to be regarded, at some concentrations, as being decidedly stimulating to some plants grown in soils, and, what is perhaps more important, relatively innocuous in large amounts. The mechanism of the stimulation obtained does not involve one single effect, but probably several. We know, for example, through experi-

<sup>35</sup> Ber. deutsch. bot. Gesell., 12, p. 8; E. S. R., 5, p. 926.

<sup>36</sup> Bot. Gaz., 27, p. 68; E. S. R., 11, p. 24.

<sup>37</sup> Bull. Soc. Vaud. Sci. Nat., 4th series, vol. 36, p. 71; Bull. Murith. Soc., Valais Sci. Nat., no. 33, p. 204.

<sup>38</sup> Ann. Agron., 27, p. 257; E. S. R., 13, p. 657.

<sup>39</sup> Staz. Sper. Agr. Ital., 40, p. 531; E. S. R., 19, p. 755.

<sup>40</sup> S. Dak. Agr. Exp. Sta., Bull. 112; E. S. R., 21, p. 436.

<sup>41</sup> Ann. Gembloux, 18, p. 609; E. S. R., 20, p. 873.

<sup>42</sup> N. Y. Agr. Exp. Sta., Bull. 41.

<sup>43</sup> Iowa Agr. Exp. Sta., Bull. 16.

ments<sup>44</sup> carried out in our laboratory, that copper is markedly effective in increasing the nitrifying activity of soils; we know, from other results which we have obtained, but not yet published, that the minerals of the soil are rendered more easily available through the action of  $\text{CuSO}_4$ ; we know from the results of Porchet and Chuard that plant cells may be directly stimulated by  $\text{CuSO}_4$ . It is therefore reasonable to explain any stimulating effects of copper in soil cultures as being of a complex nature and the results of better conditions for plant growth either directly or indirectly induced by copper through influences known to be characteristic of it as just explained.

### ZINC SULFATE

We may now review, in a manner similar to that employed for  $\text{CuSO}_4$ , the results obtained by other investigators as compared with our results on the effects of  $\text{ZnSO}_4$  on plant growth. Data indicating the toxicity of zinc to plants grown in solution cultures have been obtained by Baumann,<sup>45</sup> Jensen,<sup>46</sup> Krauch,<sup>47</sup> Storp,<sup>48</sup> True and Gies,<sup>49</sup> and Brechley.<sup>50</sup> Most of these toxic effects were obtained with relatively small quantities of zinc salts and usually under conditions antagonistic to their toxic effects owing to the presence of nutrient salts. As opposed to these evidences of the toxicity to plants of zinc, we have at times, in the work of the same investigators, manifestations of the stimulating effects of zinc in solution cultures. For example, Brechley admits in the monograph cited a slight stimulation of peas by  $\text{ZnSO}_4$ , while showing the latter to be toxic to barley. Jensen, too, whose work is described, while obtaining no stimulation for  $\text{ZnSO}_4$ , likewise showed no toxicity thereof in dilute solution, and expressed the opinion that the possibility exists of

<sup>44</sup> Lipman and Burgess, The Effects of Copper, Zinc, Lead, and Iron on Ammonification and Nitrification in Soils, Univ. Calif. Publ. Agri. Sci., vol. 1, p. 127.

<sup>45</sup> Landw. Versuchs. Stat., 31, p. 1.

<sup>46</sup> Bot. Gaz., 43, p. 11.

<sup>47</sup> Jour. für Landw., 30, p. 271.

<sup>48</sup> Landw. Jahrb., 12, p. 795.

<sup>49</sup> Bull. Torrey Bot. Club, 30, p. 390.

<sup>50</sup> Inorganic plant poisons and stimulants, Cambridge, 1914.

a stimulating power of  $\text{ZnSO}_4$  at still greater dilutions than those which he employed. More direct evidence of the stimulating effects of  $\text{ZnSO}_4$  is given by Kanda<sup>51</sup> in solutions free from nutrient salts, and by Javillier<sup>52</sup> in nutrient solutions. So far as solution cultures of all kinds are concerned, therefore, the evidence with respect to the effects of zinc on plants is conflicting, it being as strong on the side of stimulation at great dilutions of  $\text{ZnSO}_4$  as on that of lack of it or of definite toxicity.

Let us now examine the data available in which a solid substratum such as sand or soil is used instead of the solution. Direct observation of toxicity of zinc to plants in solid media is given by Storp, whose work is above cited, by Noble, Baessler, and Will,<sup>53</sup> Jensch,<sup>54</sup> Ehrenberg,<sup>55</sup> and Haselhoff and Gössel.<sup>56</sup> Evidence of the non-effectiveness of zinc either as a toxic or stimulating agent is given by Phillips,<sup>57</sup> by Holdefleiss,<sup>58</sup> and by Haselhoff and Gössel.<sup>59</sup> As against these results, however, we have many others showing definitely stimulating effects of zinc on plants grown in sand or soil. Among them may be mentioned those of Kanda,<sup>60</sup> Jensen,<sup>61</sup> Silberberg,<sup>62</sup> Zaleski and Reinhard,<sup>63</sup> Ehrenberg,<sup>64</sup> Bertrand,<sup>65</sup> Nakamura (with some plants only),<sup>66</sup> Javillier,<sup>67</sup> Roxas,<sup>68</sup> Lipman and Wilson,<sup>69</sup> and the present writers. While, however, the evidence appears to

<sup>51</sup> Jour. Col. Sci., Tokyo Imp. Univ., 19, p. 1.

<sup>52</sup> Compt. Rend., etc., 155, p. 1551.

<sup>53</sup> Landw. Versuchs. Stat., 30, p. 380.

<sup>54</sup> Ztschr. Angew. Chem., 14, p. 5.

<sup>55</sup> Chem. Zeit., 32, p. 937.

<sup>56</sup> Ztschr. Pflanzenkrank., 14, p. 193; E. S. R., 16, p. 952.

<sup>57</sup> Chem. News, 46, p. 224.

<sup>58</sup> Landw. Versuchs. Stat., 28, p. 472.

<sup>59</sup> Ztschr. Pflanzenkrank., 14, p. 193; E. S. R., 16, p. 952.

<sup>60</sup> Jour. Col. Sci., Imp. Univ. Tokyo, 19, p. 47.

<sup>61</sup> Bot. Gaz., 43, p. 11.

<sup>62</sup> Bull. Torrey Bot. Club, 36, p. 480.

<sup>63</sup> Biochem. Ztschr., 23, p. 193.

<sup>64</sup> Landw. Versuchs. Stat., 72, p. 15.

<sup>65</sup> Rev. Sci. (Paris), 49, p. 673.

<sup>66</sup> Bull. Col. Agr., Tokyo Imp. Univ., 6, p. 147.

<sup>67</sup> Ann. Inst. Pasteur, 22, p. 720; also 7th Internat. Cong. Appl. Chem., sec. vii, Agr. Chem., p. 163.

<sup>68</sup> Philippine Agric. and Forester, 1, p. 89.

<sup>69</sup> Bot. Gaz., 55, p. 409.



be overwhelmingly in favor of the stimulating effects of zinc to plant growth in soils, several instances of stimulation are qualified to hold for certain plants only or at very low concentrations of the metal. Therefore the data submitted are not as strong in favor of the stimulating effect of zinc salts to plants as one would suppose from the review above given. Nevertheless, it is strong enough in our opinion to satisfy even the critical that zinc can be a stimulant to plant growth in certain rather considerable concentrations. Besides that, its toxic effects are nowhere to be regarded as very serious if small quantities of the salt are present. Our results indicate, in addition to all this, that  $\text{ZnSO}_4$ , for example, may be stimulating to barley growth at considerable concentrations, but that the after-effects on the soil in the third season or crop may be injurious. Such injury, however, is relatively speaking, not very great unless very high concentrations of  $\text{ZnSO}_4$  are employed. Even in the third season of cropping in the case of the same soil, it appears that  $\text{ZnSO}_4$  continues to be stimulating to barley at a concentration of 200 p. p. m. of that salt as referred to the dry weight of the soil in question. Moreover, it is not unlikely that the reversal from a toxic to a stimulating condition occurring in the manganese series between the second and third crop might occur in the zinc series between the third and fourth crop. This possibility would seem to find some support from the fact that the third crop in the zinc series corresponds to the second crop of the manganese series, since two treatments—one before the first, and one before the second crop—were given to the zinc-treated pots.

#### IRON SULFATE

Results obtained in experimental trials with  $\text{FeSO}_4$  in cultures of the higher plants have been perhaps more contradictory than those noted in the cases of  $\text{CuSO}_4$  and  $\text{ZnSO}_4$  which are reviewed above. This is particularly manifest in the extensive bibliography prepared by Horton<sup>70</sup> dealing with the use of sulfate of iron in agriculture. While the latter emphasizes primarily the results obtained with  $\text{FeSO}_4$  in combating weeds,

<sup>70</sup> A Contribution to the bibliography of the use of sulfate of iron in agriculture, Chicago, 1906.

a large number of experiments are cited, among which are to be found cases of injury, ineffectiveness, and stimulation by  $\text{FeSO}_4$  to crop plants. Very few experiments appear to have been reported on the effect on plant growth of  $\text{FeSO}_4$  or other iron compounds in solution cultures. Those that are given indicate the uniformly toxic nature of iron to the higher plants under the conditions noted. For evidence on this point, the reader is referred to the investigations of Boiret and Paturel,<sup>71</sup> Gile,<sup>72</sup> Ruprecht,<sup>73</sup> Thompson,<sup>74</sup> and Knop.<sup>75</sup> No case has as yet come to our notice of the stimulating effects of iron salts to plants in solution cultures.

In soil cultures the picture is an entirely different one, and it is under those conditions that we observe the contradictory results mentioned above. Distinct cases of injury by  $\text{FeSO}_4$  to plants in soil cultures have been reported. In illustration of these, may be mentioned statements of Voelcker,<sup>76</sup> Steglich,<sup>77</sup> Nessler,<sup>78</sup> Halsted,<sup>79</sup> and others. As showing  $\text{FeSO}_4$  to be without effect on plants grown in soils, may be mentioned the experiments of Scovell and Peter,<sup>80</sup> A. Mayer,<sup>81</sup> Boiret and Paturel,<sup>82</sup> Petit,<sup>83</sup> Larbaletrier and Malpeaux,<sup>84</sup> and others. In other words, some of the investigators just mentioned, as well as Coste-Floret,<sup>85</sup> Brooks,<sup>86</sup> Griffiths,<sup>87</sup> Treboux,<sup>88</sup> and a number of others,

<sup>71</sup> *Ann. Agron.*, 18, p. 417; *E. S. R.*, 4, p. 435.

<sup>72</sup> *Jour. Agr. Res.*, 3, no. 3, p.

<sup>73</sup> *Mass. Agr. Exp. Sta.*, Bull. 161.

<sup>74</sup> *Jahresber. Agr. Chem. N. F.*, 36, p. 106.

<sup>75</sup> *Landw. Versuchs. Stat.*, 2, p. 73.

<sup>76</sup> *Jour. Roy. Agr. Soc. Eng.*, 2d ser., 1, p. 113.

<sup>77</sup> *Ztschr. Pflanzenkrankh.*, 11, p. 31; see also *Jahresber. Agr. Chem.*, 43, p. 352.

<sup>78</sup> *Centbl. Agr. Chem.*, 2, p. 125.

<sup>79</sup> *N. J. Sta.*, *Ann. Rept.*, p. 321, 1890.

<sup>80</sup> *Ky. Agr. Exp. Sta.*, Bull. 17.

<sup>81</sup> *Jour. für Landw.*, 40, p. 19.

<sup>82</sup> *Ann. Agron.*, 18, p. 417.

<sup>83</sup> *Compt. Rend.*, etc., 117, p. 1105.

<sup>84</sup> *Ann. Agron.*, 22, p. 20.

<sup>85</sup> *Prog. Agr. et Vit.*, 26, pp. 434, 463, 496.

<sup>86</sup> *Mass. Agr. Exp. Sta.*, *Ann. Rept.*, p. 42, 1896.

<sup>87</sup> *Chem. News.*, 50, p. 167.

<sup>88</sup> *Flora*, 92, p. 59.

have noted very definite stimulation of plants by  $\text{FeSO}_4$  in soil cultures. In addition to these direct results on the stimulation of plants, moreover, may be mentioned the numerous cases of stimulation of plants induced by spraying the leaves with solutions of  $\text{FeSO}_4$  either for destroying ever-present weeds in crops or for overcoming certain diseases like chlorosis. These cases are too numerous to mention here, but are well reviewed in the bibliography prepared by Horton, which is referred to above. As the discussion of our results has shown, we are in accord with the idea of the stimulating powers of  $\text{FeSO}_4$  even if used in relatively large concentrations in soils so far as the first two successive crops on the treated soil are concerned. In the third crop also, marked stimulation is obtained, but only in the higher concentrations, which in the second crop were toxic. This circumstance will be critically considered below.

#### LEAD SULFATE

The literature dealing with the subject of the effect of  $\text{PbSO}_4$ , or lead in any form on plant growth is very meager. That which is extant deals more specifically with the effect of lead sprays on foliage and fruit of trees than on the actual growth of trees, in which we are interested here. In the case of solution cultures we have found but two papers, and both of these testify to the stimulating action of  $\text{Pb}(\text{NO}_3)_2$  in dilute solutions. We refer to the investigations of Jensen<sup>89</sup> and Stoklasa.<sup>90</sup> In greater concentration the  $\text{Pb}(\text{NO}_3)_2$  was of course found to be toxic in the solution cultures. The same investigators also obtained marked manifestation of stimulation of plants in solid substrata due to lead. Jensen obtained such in quartz-sand cultures, in which greater concentrations were found stimulating than in solution cultures. Stoklasa confirmed the results of the solution cultures by field trials reported in the paper above cited, and also in other experiments<sup>91</sup> with sugar beets, oats, corn, and other crops. Voelcker<sup>92</sup> also found lead to be stimulating to wheat. When

<sup>89</sup> Bot. Gaz., 43, p. 11.

<sup>90</sup> Compt. Rend. Acad. Sci. (Paris), 156, p. 153.

<sup>91</sup> Zuckerrübenbau, 18, p. 193; E. S. R., 26, p. 225.

<sup>92</sup> Jour. Roy. Agr. Soc. Eng., 73, ent. series, 1912.

our data for  $\text{PbSO}_4$  in greenhouse soil are reviewed in the light of the foregoing, they are found to be in accord with those of Voelcker so far as the second and third crops of barley are concerned. At some concentrations in both of those series,  $\text{PbSO}_4$  acted as a stimulant to barley and often at very large or at the larger concentrations used. Our results, however, are entirely at variance with those of Jensen and Stoklasa in so far as the first crop is concerned. In that series we noted nothing but evidences of marked toxicity of the  $\text{PbSO}_4$ , with the accompanying effects on the barley plants which are described.

#### POTASH ALUM

No literature has been found on the effects on plants of potash alum in soil. The discussion set forth above giving our results with that material will therefore have to suffice.

#### MANGANESE

With the possible exception of copper, manganese and its effects on plants have received more attention at the hands of plant physiologists and students of soils than any other element here under consideration. Despite that fact, there would appear to be as much contradiction in results obtained in this case as in those of the other elements above studied. We find reports of toxicity of manganese in solution cultures in the publications of Aso,<sup>93</sup> Loew and Sawa,<sup>94</sup> and Brenchley.<sup>95</sup> On the other hand, the results of the same authors also give evidence of the stimulating effects of manganese at certain concentrations. Miss Brenchley even hints at the possibility of the existence, simultaneously, of a toxic and stimulating effect on the part of manganese and claims that either effect may show predominance, depending on the concentration of the salt employed.

On the toxic action of manganese to plants in solid substrata, principally in soils, we have the reports of experiments of

<sup>93</sup> Bull. Col. Agr., Tokyo Imp. Univ., 5, p. 177.

<sup>94</sup> Bull. Col. Agr., Tokyo Imp. Univ., 5, p. 161.

<sup>95</sup> Inorganic plant poisons and stimulants, Cambridge, 1914.

Namba,<sup>96</sup> Voelcker,<sup>97</sup> Kelley,<sup>98</sup> and Guthrie and Cohen.<sup>99</sup> As opposed to these, however, we have numerous cases on record of the stimulating effects of manganese to plants grown in soil, and even the work of the investigators last named is by no means to be considered as absolute evidence against such action of manganese, since the toxic action observed was in some cases very slight, and some of the concentrations involved were so unusually high as to leave little expectation of anything but toxicity of manganese for the plants tested. Among the investigators referred to who have furnished evidence of the stimulating effects of manganese, may be mentioned Voelcker,<sup>100</sup> Bertrand,<sup>101</sup> Roxas,<sup>102</sup> Loew and Sawa,<sup>103</sup> Nagaoka,<sup>104</sup> Loew and Honda,<sup>105</sup> Fukutoma,<sup>106</sup> Namba,<sup>107</sup> Uchiyama,<sup>108</sup> Takeuchi,<sup>109</sup> Feilitzen,<sup>110</sup> Strampelli,<sup>111</sup> and Lipman and Wilson.<sup>112</sup> While in this review we have omitted a number of the investigations bearing on the subject, enough have been given to indicate clearly the trend of the evidence in hand. Fuller bibliographies may be obtained in the excellent reviews of the literature given by Brenchley<sup>113</sup> and by Kelley.<sup>114</sup>

Comparing the results of other investigators with ours, some interesting differences, as well as similarities, between them become evident. For example, we are in accord with most of the investigations above reviewed as favoring the existence of stimu-

<sup>96</sup> Bull. Col. Agr., Tokyo Imp. Univ., 7, p. 635.

<sup>97</sup> Jour. Roy. Agr. Soc. Eng., 64, p. 348.

<sup>98</sup> Jour. Ind. Eng. Chem., 1, p. 533.

<sup>99</sup> Agr. Gaz. N. S. Wales, 21, p. 219.

<sup>100</sup> Jour. Roy. Agr. Soc. Eng., 44, p. 348.

<sup>101</sup> Compt. Rend., etc., 124, p. 1032.

<sup>102</sup> Philippine Agr. and Forester, 1, p. 89.

<sup>103</sup> Flora, 91, p. 264.

<sup>104</sup> Bull. Col. Agr., Tokyo Imp. Univ., 5, p. 467; 6, p. 135.

<sup>105</sup> Bull. Col. Agr., Tokyo Imp. Univ., p. 6. 125.

<sup>106</sup> Bull. Col. Agr., Tokyo Imp. Univ., 6, p. 137.

<sup>107</sup> Bull. Col. Agr., Tokyo Imp. Univ., 7, p. 635.

<sup>108</sup> Bull. Imp. Cent. Agric. Exp. Sta., Japan, 1, p. 37.

<sup>109</sup> Jour. Col. Agr., Tokyo Imp. Univ., 1, p. 207.

<sup>110</sup> Jour. für Landw., 55, p. 289.

<sup>111</sup> 6<sup>o</sup> Cong. Internat. Chem. Appl. Roma, 4, p. 14.

<sup>112</sup> Bot. Gaz., 55, p. 409.

<sup>113</sup> Inorganic plant poisons and stimulants, Cambridge, 1914.

<sup>114</sup> Hawaii Agr. Exp. Sta., Bull. no. 26.

lation by manganese of the growth of barley so far as the first crop on the soil in question is concerned. In the case of the second crop, however, a depression in yield of considerable magnitude is induced by  $\text{MnSO}_4$  and a stimulation produced by  $\text{MnCl}_2$  in the higher concentrations of the salt, while the lower ones depress the yield like  $\text{MnSO}_4$ . In the third crop, as we have already seen, there is a practical disappearance of all toxic effects in both of the manganese series which we had under observation, and taking the place of the former toxic effects we find marked stimulating effects. The indication is therefore that in general our results are in accord with those of the investigators cited above who attributed to manganese stimulating effects for plants.

## ADDITIONAL INVESTIGATIONS

### NITRIFICATION

Earlier experiments by P. S. Burgess<sup>115</sup> and the senior author had demonstrated the stimulating effects of  $\text{CuSO}_4$ ,  $\text{FeSO}_4$ ,  $\text{ZnSO}_4$ , and  $\text{PbSO}_4$  on nitrification in soils. We were therefore led to wonder whether much, if not all, of the stimulation exerted on the higher plants by most of the salts in the first crop was due to the increase in the available supply of nitrogen there through the effects of the salts. Accordingly, tests of the nitrifying powers of the soils in a number of the pots in every series were made by the usual laboratory methods employed for such purposes. Dried-blood nitrogen was used as the nitrifiable material at the rate of 1 per cent of the dry weight of the greenhouse soil. Lack of space forbids the presentation here of the large amount of data collected on the subject now under consideration. We may, however, refer to the striking features thereof, owing to their undoubted connection with the cause or causes of the stimulating effects above noted. In the second crop of the copper series in the greenhouse soil the nitrifying power was from 10 per cent to 50 per cent greater in the "coppered" than in the "uncoppered" soil. In the third crop, which, it

<sup>115</sup> Univ. Calif. Publ. Agr. Sci., 1, p. 127.

will be remembered, was grown one year after the second and last copper application had been made, the increases in the nitrifying powers of the treated soils were from 33 per cent to 100 per cent greater than in the control soils receiving no copper. In a general way the highest concentrations of  $\text{CuSO}_4$  gave the largest increases in nitrifying power in the second crop, but in the third crop there was more or less irregularity in that regard and the smaller concentrations appeared to be as effective as the larger.

In the case of the zinc series, determinations of the nitrifying powers of the different soils were made after the third crop only. In that case also, the nitrifying power was increased by applications of  $\text{ZnSO}_4$  equivalent to 200, 600, and 1000 p. p. m. The increases, however, were much smaller than in the case of the  $\text{CuSO}_4$  and varied from 3 per cent to 16 per cent at the different concentrations, the most favorable concentration being 600 p. p. m. An important difference exists between the  $\text{CuSO}_4$  and the  $\text{ZnSO}_4$  series in that all the concentrations of the former which were employed increased the nitrifying powers of the soil in the third crop to some extent, while only the concentrations just given were instrumental in imparting such a stimulus in the case of the latter salt. Iron behaved very similarly to zinc in most respects so far as the soil's nitrifying powers were concerned, and 0.2 per cent, 0.4 per cent, 0.6 per cent, and 0.8 per cent were the range of concentration of  $\text{FeSO}_4$  corresponding to those named for  $\text{ZnSO}_4$  above. One difference between iron and zinc in their influences on nitrification in the greenhouse soil is that the former does not seem to have been appreciably toxic in any concentration, even though as much as 2 per cent  $\text{FeSO}_4$  was employed, whereas the latter, as we have already seen, markedly depressed the soil's nitrifying power when used in excess of 0.1 per cent of the dry weight of the soil. Like  $\text{ZnSO}_4$  and  $\text{FeSO}_4$ ,  $\text{PbSO}_4$  was tested as to its effect on nitrification after the third crop only. Under those conditions it gave, however, very different results from the other salts, since no stimulation to nitrification was noted at all, no matter what amounts of  $\text{PbSO}_4$  were employed. On the other hand, while  $\text{PbSO}_4$  was throughout slightly toxic to nitrification under the

conditions named, the toxicity seemed to be about the same with the larger as with the smaller concentrations of  $\text{PbSO}_4$  employed.

The manganese salts were tested in the first crop only, in connection with their powers to affect nitrification. The following were the results:  $\text{MnSO}_4$  was not toxic under the conditions named in any of the concentrations in which it was employed, 0.6 per cent being the highest. It appeared to be very slightly stimulating at all concentrations. In the case of  $\text{MnCl}_2$  we find marked toxicity to nitrification at concentrations in excess of 0.4 per cent, and very distinctly toxic effects at concentrations in excess of 0.15 per cent. On the other hand, we also note that nitrification was stimulated by the following concentrations: 0.05 per cent, 0.1 per cent, and 0.15 per cent. The stimulation was very marked only in the case of the latter two concentrations and was very much in excess of that induced by  $\text{MnSO}_4$  at any concentration.

The nitrifying powers of the Oakley blow sand employed in one copper series, which is described above, were also determined. Marked stimulation to the nitrifying power of the soil was noted at concentrations of  $\text{CuSO}_4$  equivalent to 100 p. p. m., 200 p. p. m., and 300 p. p. m., the first two being most marked. Ammonium sulfate was employed as the nitrifiable material. Amounts of  $\text{CuSO}_4$  in excess of 300 p. p. m. were decidedly toxic, and very little or no nitrification occurred in the soil containing more than 700 p. p. m.  $\text{CuSO}_4$ .

While there is considerable discrepancy in the correlation of the effects of the different salts on barley growth and on the nitrifying bacteria, there appears to be a general relation, at least, between the stimulating effect exerted by a salt on the nitrifying flora and its effect on the barley plant. The serious irregularities which seem to militate at present against the definite establishment of such a relationship based on our data can undoubtedly be explained on the basis of certain factors like the residual nitrate supply in soils and the differences in its distribution throughout the soil mass which of course must exist. While therefore we make no attempt to assert that the stimulating effects and perhaps the toxic effects to barley exhibited by



the salts here under discussion are to be accounted for by their effects on the nitrifying flora and hence on the available nitrogen supply, we do believe that the latter is one of the few important factors—perhaps the most important—involved in the problem of explaining stimulation of plants in soils particularly, and possibly also, to some extent, the toxicity of salts in soils. That the effects of the salts on the nitrifying powers of the soils here studied are not the exclusive cause of the phenomena above discussed, we can probably believe with confidence. The total quantities of citric acid-soluble phosphoric acid and potash in soils have been found by us to be augmented through the action of the metallic sulfates in question, and we are also aware of the possible inhibiting effects of those salts for certain factors which may be inimical to the proper development of the soil bacteria.

In addition, there can be no question about the profundity of the changes in the soil's physical condition induced by any metallic sulfate and about the effects which follow in its wake. Most notable of all facts in that connection is the fluffy and pulverulent condition of the soils treated with ferrous sulfate, due undoubtedly to the formation of hydrated ferric oxide and other similar compounds. Special studies (unpublished) carried out by Mr. H. H. Coolidge on the soils of the ferrous-sulfate series, showed that the treatment of the soil reduced its power to raise water to a certain point, while at first allowing it to raise it faster; that the hygroscopicity of the soil was reduced; that its total water-holding power and its water-retentiveness were diminished; that its percolation power was increased; that its moisture-equivalent was diminished. Mr. Coolidge also found that, contrary to the effects of  $\text{CuSO}_4$  and some of the other sulfates, the soil's water-soluble phosphorus and potassium were very much reduced in quantity by treatment with  $\text{FeSO}_4$ . Different and numerous though these effects be, there can be little question that they must influence, to some degree at least, the soil's nitrifying power. A further discussion of this phase of the problem is, however, impossible at this time and must await consideration in connection with some of our other studies.

## NITROGEN CONTENT OF THE GRAIN

It appeared of interest, in view of the foregoing, to determine to what extent the soil's nitrate content, which was high throughout, had influenced the nitrogen content of the dry matter. We therefore determined the nitrogen content of the grain harvested in a number of the series so as to obtain some idea of the direction taken by the effects of the nitrates, if any were exerted. As a result of these analyses it was found that in the second crop of the copper series the nitrogen content of the grain was in the absolute from 0.14 per cent to 0.57 per cent higher in the case of that grown on the "coppered" soil than in that grown on the control soils. In the third crop of the copper series the nitrogen content was from 0.05 per cent to 0.38 per cent higher in the grain from the treated soils than in that from the untreated soils. In the case of the second crop of the zinc series, the nitrogen content of the grain was from 0.06 per cent to 0.64 per cent higher in the grain of the treated than in that of the untreated soils. In the third crop of the zinc series, the corresponding figures ranged from nothing in one case, in which the lowest concentration of  $\text{ZnSO}_4$  was used, to 0.42 per cent. Similarly in the case of the iron series, the range was from nothing to 0.68 per cent in the second crop, and from 0.22 per cent to 0.50 in the third crop. No determinations were made in the case of the lead series, but analyses were carried out in the case of the second crop of the potash alum series which indicated that the grain of the treated soils was in most cases only very slightly richer in nitrogen than that from the untreated soils, and that the maximum increase did not surpass 0.09 per cent.

On the whole, and leaving the potash alum out of consideration, it seems that one of the results of stimulation of the barley plant by the metallic sulfates in question was the increase in the nitrogen content of the grain. At all concentrations of all the salts tested, with only one or two exceptions, the grain grown on the treated soils was richer in nitrogen than that on the untreated soils. That this fact should be referable primarily to the increased vigor of the nitrate formation in the treated

soils induced by the presence of the salts appears to the writers rational and justifiable. However that may be, there can be no question that even in the second and third crops on the soil under examination the nitrogen content of the grain shows its superiority in the case of the treated soils as against the untreated soils. If this should prove true on soils in general, and there is strong likelihood that it will, should it not offer us a method for increasing the nitrogen content of our grain, a problem which has for some time been agitating agronomists and flour producers in California? While, as has been indicated by other investigators, a high nitrogen content of grain may not necessarily imply a high gluten content of the flour, the latter being the consummation anxiously sought, it is at least likely that the generally higher nitrogen content of grain will also bring with it a higher gluten content. Since, moreover, our investigations indicate that small quantities of the metallic salts are as effective in inducing the enrichment of grain in nitrogen as the larger quantities, it is further possible that the means suggested of raising the gluten content of grain may prove to be a very inexpensive one.

#### ABSORPTION OF METALS BY SOIL AND PLANT

In discussing such problems as the one which forms the subject here, the technical chemist will frequently ask to what extent plants will absorb such metals as have been studied by us. The literature on that topic is so rich in evidence that metals are readily absorbed, and in considerable quantity, by the plant that we did not deem it desirable to go at length into such an investigation with our harvested barley plants as a basis. We did, however, make analyses of a number of plants from pots receiving different treatments and also of the soils in some of the pots. We are therefore in a position to answer partly on the basis of our own data, the question above raised. On the subject of the absorption of metals by plants, the reader is referred for full and interesting discussions to Czapek,<sup>116</sup>

<sup>116</sup> *Biochemie der Pflanzen*, Jena, 1905.

Pfeffer,<sup>117</sup> Müller,<sup>118</sup> Lehmann,<sup>119</sup> and Brenchley.<sup>120</sup> In our analyses both grain and straw were examined, and copper and zinc only were determined. These were both determined electrolytically. Unlike Vedrodi,<sup>121</sup> we could find nothing more than traces of copper or zinc in the grain, but succeeded easily in obtaining definite quantities of those metals in the straw from some of the pots. In the first crop, from thirty-six to forty-three grams of straw were taken for analysis for copper, and, after ashing, the mineral residue was prepared for analysis for copper by the method above mentioned, straw from the pots receiving 100, 800, 1100, and 1200 p. p. m.  $\text{CuSO}_4$  being employed. In the first case, the percentage of copper in the straw varied from nothing to 0.0006 per cent. In the second case, the percentage of copper was 0.0002. In the third case, it was 0.0033 per cent, and in the fourth case, 0.0044 per cent.

In the pots receiving  $\text{ZnSO}_4$ , there were chosen for analysis the straw produced in those receiving 100, 300, 500, 1000, 1100, 1200, 1300, and 1600 p. p. m. In the first case, the analysis showed the presence of zinc to the extent of 0.00036 per cent; in the second, 0.0008 per cent; in the third, 0.003 per cent; in the fourth, 0.017 per cent; in the fifth, 0.013 per cent; in the sixth, 0.013 per cent; in the seventh, 0.01 per cent; and in the eighth, 0.012 per cent. In the cases of both zinc and copper the percentage of the metals absorbed by the barley plant was smaller than that reported as being absorbed by the plants studied by other investigators whose work is referred to in the literature last cited. In general, it seems that up to a certain point increasing quantities of the metals added to the soil induce larger absorptions of metal by the plant, but beyond that point the addition of metals to the soil appears to be without effect in inducing further absorption. This seems to be particularly true in the case of the zinc. We do not desire, however, to draw any conclusions from the relatively meager data which we have gathered on the subject in question under

<sup>117</sup> *Pflanzenphysiologie*, Leipzig, 1897 and 1901.

<sup>118</sup> *Ztschr. Pflanzenkrankh.*, 4, p. 142.

<sup>119</sup> *Arch. Hyg.*, 27, p. 1.

<sup>120</sup> *Inorganic plant poisons and stimulants*, Cambridge, 1914.

<sup>121</sup> *Chem. Ztg.*, 20, p. 399.

the conditions here discussed. Since it is rare in nature that more than the lowest concentrations of copper and zinc here studied ever occur in soils suited to crop production, the question of the danger in the use by man and animals of plants absorbing copper is not a serious one, for with small quantities of copper and zinc present in the soil, very small quantities only are absorbed by the plant. It must be added here, moreover, that we employed easily water-soluble salts, whereas in nature the compounds of the metals found are principally those of a very insoluble nature. The latter circumstance would perforce make impossible any large concentration of any metal in the soil solution, and hence only small quantities could be absorbed by plants.

We were interested also in obtaining an inkling as to the fate of the copper and zinc added to the soil after three seasons of plant growth thereon. Accordingly, several soils were chosen for examination. Pots receiving 600, 1800, 2000, and 3000 p. p. m.  $\text{CuSO}_4$  gave the following results: In the first case all the copper added was recovered. In the second case 1750 p. p. m., instead of 1800, were recovered. In the third case all the copper was recovered, and in the fourth case 2875 p. p. m. were recovered, instead of 3000 p. p. m.

In the case of zinc the pots receiving 800, 1700, and 2000 p. p. m.  $\text{ZnSO}_4$  were studied. In the first case 750 p. p. m. were recovered. In the second case 1650 p. p. m. were recovered in one soil, and 1500 p. p. m. in another soil. In the third case only 1250 p. p. m. were recovered.

These data indicate that in the case of copper, at least, the soil clings tenaciously to the metal; and most of it, or nearly all of it, can be recovered from the soil even three seasons after it has been incorporated therewith, and three crops of barley grown in the interim. With zinc, there do appear to be losses. These may perhaps be explained in part by the larger amounts of zinc than copper absorbed by plants, and by the lesser accuracy of the method for its determination as compared with that employed for copper. Twenty-gram samples of soil were employed in all cases for obtaining the extracts which were analyzed, and it is therefore believed that the error involved in the analyses could not have been very large.

## GENERAL AND PRACTICAL CONSIDERATIONS

The practical as much as the theoretical point of view inspired these investigations. In a time such as this, when the smelter question is of great significance in agricultural districts and when outcries against the damage caused by both smelter fumes and solid smelter wastes are most insistent, it appeared to us that the moment had arrived for wholly disinterested investigators to examine into it. Our experiments as described in this paper have dealt, in the main, with the effects on barley growth in three successive crops of the metals which would be likely to be deposited in the vicinity of smelters and gradually washed down into sources of irrigation water for the territory lying below the smelter plants. Despite the fact that we have used much more soluble forms of the so-called toxic salts than are likely to occur under the conditions just described, and despite the fact that we have employed both large and small amounts of these salts, we are unable to read into our results any serious danger to agriculture from the solids of smelter wastes as they may be transported to cropped lands by irrigation water. In making this statement we are not unmindful that very small areas occur<sup>122</sup> near the smelter plants in which the tailings may be carried down by streams and deposited on land in large quantities. These may, for example, carry enough of the toxic heavy metals to render land poor in producing capacity. But in the first place the most prejudiced persons will not claim that such affected areas of agricultural land are more than negligible quantities when the question is considered in the large; in the second place, even under conditions so extreme, none but the most biased will deny that proper methods of management can be made to render innocuous any harmful effects which the tailings in question may be potentially capable of exerting. These methods of management are clearly indicated and include the impounding of water carrying tailings or the passage of stream water through screens which will separate out

<sup>122</sup> See R. H. Forbes, "Certain effects under irrigation of copper compounds upon crops," *Univ. Calif. Publ. Agri. Sci.*, 1, no. 12, 1917.

the tailings, and, in the case of the land which is already affected, the use of organic matter. The indications of our experiments are that a year or two of fallowing will usually correct the difficulty.

We are therefore obliged to reaffirm the position taken by Lipman and Wilson<sup>123</sup> to the effect that there seems to be little danger in store for our agricultural lands in the metallic residues which are deposited by smelters in their vicinity and from their ultimate solution in small degree in potential irrigation water-supplies which may be subsequently transported to farm lands. On the contrary, we give evidence above that so far from being toxic to barley plants, small amounts of the metals studied may be distinctly stimulating to them. While this is more strictly true in the case of some metals than of others, it appears none the less to be so. Moreover, in cases in which toxicity is effected by the application of any of the metallic sulfates named, it is usually very slight, even when large quantities of the salts are employed. While we have experimented in this series of investigations only with barley, evidences given by ourselves and by others who are above cited, indicate that a number of other plants behave similarly to barley, if not exactly like it. From the practical standpoint, therefore, we cannot see that any other conclusion can be reached than that we may virtually ignore any deleterious effects which may be urged against the metals of smelter wastes which are here discussed. We use the word "practical" here advisedly, because if solution instead of soil cultures were taken as a criterion, our standard of judgment would not be practical. Whatever may be said about soil cultures, one must admit that they approximate most closely of any greenhouse or laboratory methods the natural conditions under which crops grow. We cannot see that any other culture than one which at least offers a solid substratum to the plant may be regarded as valid in the determination of whether or not salts like those here under consideration are, under the conditions of the smelter and its vicinity, a menace to plant growth.

<sup>123</sup> Bot. Gaz., 55, no. 6, p. 409. June, 1913.

To all this, however, there must be added some other considerations. One of them serves to qualify, in some measure at least, the remarks made above, and the other to supplement them. While the metals studied by us do not seem to have given evidence, under the conditions of our experiment, of any serious injury to barley, a non-metal, arsenic, has given marked evidences of toxicity to barley under similar conditions. Arsenic, being found frequently in conjunction with the other elements in the vicinity of smelters, is necessarily a subject worthy of attention. Our results with its use in soil cultures are not yet ready to be reported, but we hope sometime in the near future to publish them. Suffice it to say now that such compounds of arsenic as arsenic trisulfide and Paris green have proved to be extremely toxic to barley in both heavy and light soils, while lead arsenate has proved to be only slightly toxic. Whether or not arsenic oxide, which is the form to be expected in lands in the vicinity of smelters, will act similarly remains to be shown by further experiments which are now being planned by us.

We are constrained to add to the foregoing that we have borne in mind the difference in the effects produced on a toxic material by the change in a soil's constitution. Indeed, our experiments with copper in three widely different types of soil testify to that fact; and while we have found marked differences in the degrees of stimulation and toxicity of copper in the different soil types, all of the latter appear to have given both stimulation and toxicity. Even in the sandy soil in which the toxicity of  $\text{CuSO}_4$  became manifest at the lowest concentration for any of the types of soil studied, as much as 0.03 per cent of  $\text{CuSO}_4$  of the dry weight of the soil still acted as a stimulant to barley. Considering that  $\text{CuSO}_4$  is an easily water-soluble salt, it would be reasonable to expect that such compounds as  $\text{Cu}(\text{OH})_2$ ,  $\text{CuCO}_3$ , which are the usual forms to be expected in soils near smelters, could be tolerated by plants in much larger quantities.

If, as appears to us reasonable, we should be able to accept the data above offered by us, at least as tentative evidence that we have little to fear from the solids of smelter wastes in the contamination of our irrigation water-supply and therefore in



injuring large areas of land, we have another interesting proposition to bring forward. L. T. Sharp and the senior author<sup>124</sup> have already reported preliminary pot experiments in evidence of the fact that  $\text{H}_2\text{SO}_4$  exerts a remarkable effect on alkali soil with the result of changing the latter from an unproductive state to a productive one. The probable reasons for this action are discussed in the paper referred to above. Suffice it to say here that field experiments which still remain unpublished amply confirm the pot experiments. If this should prove to be a more or less permanent effect on alkali soils which do not contain too high a percentage of salts (from 0.6 per cent to 0.8 per cent), then we could solve the other and really serious phase of the smelter question, namely, the smelter gases. Chemical engineers of note, including F. G. Cottrell of the Bureau of Mines, have often stated to the senior author that the chief reason that  $\text{SO}_2$  fumes from the smelters are not made into  $\text{H}_2\text{SO}_4$  is because there would be no use for such tremendous quantities of that acid. If, however, we should be able to apply  $\text{H}_2\text{SO}_4$  to many alkali soils with good effect that objection would vanish. If, therefore, the smelters will only produce the acid cheaply enough, as they now seem inclined to do, we shall be able to banish much costly litigation, let the smelter industry develop untrammelled, give the smelter companies compensation for oxidizing the  $\text{SO}_2$ , and last but not least, put large acreages of barren land into good crop-producing condition.

This proposition sounds almost chimerical, but much thought and work on it have convinced us that it is well justified by facts, and we believe that the condition just described will speedily come to pass. We mention the  $\text{SO}_2$  problem here only in passing, since much fuller discussion of our experiments with  $\text{H}_2\text{SO}_4$  on alkali soils is to appear in later papers. Suffice it to say, that we believe we have in it and in the experiments above discussed strong evidence of methods for controlling the smelter nuisance without injuring the industry or the farmer, and, besides, much evidence on the true effects of solids of smelter wastes on barley grown in soils.

<sup>124</sup> Univ. Cal. Publ. Agri. Sci., vol. 1, p. 275.

## THEORETICAL CONSIDERATIONS

A few words may not be out of place here with regard to the mechanism of the action of the different salts employed in our experiments, be such action in the direction of stimulation or in that of toxicity. In the first place, the salts in question must exercise some effect on the cell of the root itself and, through it, on the whole plant. If this were not so, we should not obtain the stimulating as well as the toxic effects of a given salt in solution cultures, as well as in soil cultures. In the latter, we do of course obtain more definite evidence of stimulation than in the former, and for that reason we may claim with some justice, as we have above, that stimulation effects are chiefly attributable to some influence, not always the same, induced by the salt on the soil, rather than on the root of the plant. This does not, to be sure, deny the existence of the latter effect in soil cultures and particularly in solution cultures; but when the most marked stimulation occurs, it is rarely noted in the latter. We therefore believe it reasonable to suppose that we are dealing under such circumstances with an effect on the soil, rather than with one on the plant root. What such salt effects on the soil may be like are explained above. It is not easy, however, to explain or even to speculate on an explanation of the effect of a salt directly on the plant root in the direction of stimulation. We have no unexceptionable evidence on the subject of compounds of copper, for example, with albuminoid material of living cells, and that increases the difficulty of accounting for observed facts of stimulation. It is nevertheless possible that stimulation of root cells by copper may be due to an effect of the latter in decreasing or increasing the permeability of the cell, or perhaps to the possible small content of iron in the copper compounds employed, the iron acting as one of the essential elements to cell development. Neither of these speculations at present appears to have value other than that of inducing further thought and discussion on the subject. So far as the toxic effects of salts on plants in solution cultures is concerned, nothing need be added here to the excellent discussions already given by Czapek and Pfeffer which are cited above, and by Hober.<sup>125</sup>

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<sup>125</sup> *Physikalische Chemie der Zelle und der Gewebe*, Leipzig, 1914.

With regard to stimulation in soil cultures, there may be added here something which is not mentioned in the discussion above, namely, that the salts of the heavy metals may act with respect to oxydases as Loew<sup>120</sup> has claimed manganese does, augmenting their activity and thus preventing the accumulation of toxic materials in the soil. That such a catalytic effect does exist is, however, very doubtful in the light of present evidence. That other forms of catalytic effects may be exerted by such salts as those employed in our experiments is at least not impossible.

### SUMMARY

The authors have been carrying on a series of investigations on the effects of  $\text{CuSO}_4$ ,  $\text{ZnSO}_4$ ,  $\text{FeSO}_4$ ,  $\text{PbSO}_4$ ,  $\text{MnSO}_4$ ,  $\text{MnCl}_2$ ,  $\text{KAl}(\text{SO}_4)_2 \cdot 12 \text{H}_2\text{O}$ , and different forms of arsenic on the growth of barley. The experiments were carried out in paraffined earthenware pots nine inches in diameter, greenhouse soil made up from clay adobe soil and barnyard manure being used principally. In the case of  $\text{CuSO}_4$ , two other soils were used in addition to the greenhouse soil, namely, the Oakley blow sand and the Berkeley clay adobe. With the greenhouse soil the experiment continued for three successive crops of barley; with the clay adobe soil, for two crops; and with the blow sand for only one crop. The results of these experiments, which are set forth in the tables and discussion above, may be summarized and their significance indicated briefly as follows:

1. In the greenhouse soil, in the first crop  $\text{CuSO}_4$  acts as a stimulant throughout from concentrations of 50 p. p. m. to 600 p. p. m. inclusive. When the roots are left out of consideration, it acts as a stimulant even to the highest concentration employed, viz., 1500 p. p. m.

In the second crop  $\text{CuSO}_4$  acts as a stimulant to both roots and tops up to and including 1800 p. p. m., and is without effect on the roots, while stimulating to tops even at 2800 p. p. m. Grain production is stimulated by  $\text{CuSO}_4$  in the second crop practically throughout the series.

<sup>120</sup> *Flora*, 91, p. 264.

In the third crop both root and top production are stimulated up to and including concentrations of  $\text{CuSO}_4$  equivalent to 2200 p. p. m. Grain production is almost similarly stimulated.

2. In the clay adobe soil in the first crop straw, grain, and root production are all stimulated up to and including concentrations of  $\text{CuSO}_4$  equivalent to 800 p. p. m.

In the second crop no stimulation takes place in the 100 and 200 p. p. m. concentrations, but in all higher concentrations, at least including that equivalent to 900 p. p. m. This holds for both straw and root production.

3. In the Oakley blow sand, only one crop being grown,  $\text{CuSO}_4$  stimulates markedly grain production and slightly straw and root production at concentrations up to and including 300 p. p. m.  $\text{CuSO}_4$ .

4. In the greenhouse soil in the  $\text{ZnSO}_4$  series the first crop is stimulated both as to root and straw yields throughout at concentrations varying from 100 p. p. m. to 2000 p. p. m.  $\text{ZnSO}_4$ .

In the second crop stimulation to straw and root yields occurs at 200 p. p. m.  $\text{ZnSO}_4$ , and marked stimulation to root yield without effect on straw yields up to 600 p. p. m.  $\text{ZnSO}_4$ . Beyond that point slight toxicity sets in and is maintained almost uniformly throughout.

In the third crop neither stimulation nor toxicity is apparent at concentrations of 200 p. p. m.  $\text{ZnSO}_4$ , but concentrations in excess of the latter are distinctly toxic.

5. In the greenhouse soil in the  $\text{FeSO}_4$  series, the first crop shows the stimulating effects of  $\text{FeSO}_4$  throughout in concentrations varying from 0.1 per cent to 1 per cent. The straw yields are increased throughout and the root yields slightly so up to and including the concentration 0.7 per cent  $\text{FeSO}_4$ .

In the second crop  $\text{FeSO}_4$  stimulates straw production in concentrations varying from 0.2 per cent to 1 per cent inclusive. Grain production is only slightly and irregularly stimulated at the same concentration. Root production is affected similarly to the grain production.

In the third crop concentrations from 1 per cent  $\text{FeSO}_4$  up to and including 2 per cent are markedly stimulating to straw and grain yields and very slightly effective in both directions in

regard to root yields. Smaller concentrations than those mentioned slightly depress straw and root production, but definitely stimulate grain production.

6. In the greenhouse soil in the  $\text{PbSO}_4$  series, first crop, the straw production is depressed by about one-third the total amount produced in the control. The depression appears to be uniform at concentrations of from 200 p. p. m. to 1500 p. p. m.  $\text{PbSO}_4$ . Likewise, the root yields are depressed by even a greater figure (about 60 per cent), and again almost uniformly throughout.

In the second crop the straw production is nowhere depressed in the entire series and is stimulated at concentrations of from 300 p. p. m. to 600 p. p. m.  $\text{PbSO}_4$  as well as at scattering concentrations in excess. Root production, on the other hand, is slightly depressed throughout.

In the third crop the straw production is markedly stimulated at concentrations varying from 1000 p. p. m. to 2400 p. p. m.  $\text{PbSO}_4$ , but slightly depressed at lower concentrations. Grain production is similarly affected, and the  $\text{PbSO}_4$  remains without effect on the roots within the same limits of concentration.

7. In the greenhouse soil in the potash alum series the first crop shows stimulation to straw yields at all concentrations varying from applications of 300 pounds to 2000 pounds  $\text{K}_2\text{O}$  per acre. Root yields are stimulated at the lowest concentration named, but scarcely at all in the others.

In the second crop the straw yields are again stimulated by the doubling of the potash alum application throughout the series. Relatively the stimulation is much greater than in the first crop. Grain production and root production are also markedly stimulated, the former at the smaller applications of potash alum and in other isolated instances, and the latter throughout.

In the third crop the straw yield is markedly depressed throughout. The grain yields are slightly stimulated in some cases, and in the balance remain unaffected. The root yields are depressed similarly to the straw yields.

8. In the greenhouse soil in the  $\text{MnSO}_4$  series the first crop is stimulated in regard to straw yields at all concentrations be-

tween 500 p. p. m. and 3000 p. p. m.  $\text{MnSO}_4$ , but most markedly at 1500 p. p. m. The root yields are also markedly stimulated, but only at concentrations up to and including 1500 p. p. m. Beyond that concentration, root yields are more or less reduced.

In the second crop the straw yields are stimulated at from 4000 p. p. m. to 6000 p. p. m.  $\text{MnSO}_4$ , but markedly depressed at concentrations below 4000 p. p. m. The grain yields are about equally depressed throughout, but not markedly. The root yields are depressed throughout the series rather markedly, the smallest depression occurring at concentrations of 2000 and 2500 p. p. m.  $\text{MnSO}_4$ .

In the third crop a stimulation is induced toward the production of straw, grain, and roots, the medium concentrations being most effective. Little or no evidence of toxic effects of  $\text{MnSO}_4$  was observed.

9. In the greenhouse soil in the  $\text{MnCl}_2$  series the first crop is markedly stimulated in straw production at concentrations varying from 500 to 1500 p. p. m.  $\text{MnCl}_2$ . Beyond the latter concentration,  $\text{MnCl}_2$  becomes more and more acutely toxic, until almost no straw is produced at 6000 p. p. m.  $\text{MnCl}_2$ . Root production is affected similarly to straw production, in a general way.

In the second crop straw production is stimulated throughout except at the two lowest concentrations—500 and 1000 p. p. m. respectively. Grain yields, however, are depressed almost throughout. The depression is relatively slight (there being one case of stimulation) at concentrations varying from 500 p. p. m. to 3000 p. p. m. Above the latter concentration, the  $\text{MnCl}_2$  is markedly toxic to grain production. Root production is markedly depressed throughout.

Like  $\text{MnSO}_4$ ,  $\text{MnCl}_2$  exerts a stimulating effect on the yields of straw, grain, and roots in the third crop. Again, little or no evidence of a toxic effect was noted in this series.

10. Results are given on the effect of the salts used on the nitrogen content of the grain produced, on the nitrifying powers of the soils concerned, on the amounts of copper and zinc taken up by some of the barley plants in the different series; and

also correlations are drawn between some of these factors and the complete yields of dry matter.

11. Some practical and theoretical phases of the smelter question are discussed, and the evidence above given is employed to show that from the large practical standpoint the solids of smelter wastes cannot justly be considered a menace to agriculture.

12. Many other points of interest are discussed in connection with the smelter problem as a whole and with the results of our experiments.

*Transmitted September 7, 1916.*

TABLE IIa  
CuSO<sub>4</sub> SET—FIRST CROP—GREENHOUSE SOIL

	CuSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	50	47.8	42.80	.....	.....	47.8	42.80	10.2	9.05	58.0	51.85	+12.09
2	50	37.8		.....	.....	37.8		7.9		45.7		
3	100	43.5	37.75	.....	.....	43.5	37.75	8.7	9.45	52.2	47.20	+ 7.44
4	100	32.0		.....	.....	32.0		10.2		42.2		
5	200	47.9	42.20	.....	.....	47.9	42.20	10.0	8.25	57.9	50.45	+10.69
6	200	36.5		.....	.....	36.5		6.5		43.0		
7	300	41.6	40.55	.....	.....	41.6	40.55	9.5	10.75	51.1	51.30	+11.54
8	300	39.5		.....	.....	39.5		12.0		51.5		
9	400	37.2	39.00	.....	.....	37.2	39.00	7.9	7.85	45.1	46.85	+ 7.09
10	400	40.8		.....	.....	40.8		7.8		48.6		
11	500	38.2	36.70	.....	.....	38.2	36.70	9.8	8.50	48.0	45.20	+ 5.44
12	500	35.2		.....	.....	35.2		7.2		42.4		
13	600	46.5	47.00	.....	.....	46.5	47.00	8.2	8.20	54.7	55.20	+15.44
14	600	47.5		.....	.....	47.5		8.2		55.7		
15	700	38.7	39.85	.....	.....	38.7	39.85	7.0	6.25	45.7	46.10	+ 6.34
16	700	41.0		.....	.....	41.0		5.5		46.5		
17	800	50.7	45.35	.....	.....	50.7	45.35	7.2	6.45	57.9	51.80	+12.04
18	800	40.0		.....	.....	40.0		5.7		45.7		
19	900	51.2	45.70	.....	.....	51.2	45.70	4.8	5.35	56.0	51.05	+11.29
20	900	40.2		.....	.....	40.2		5.9		46.1		
21	1000	44.6	43.55	.....	.....	44.6	43.55	5.5	5.25	50.1	48.80	+ 9.04
22	1000	42.5		.....	.....	42.5		5.0		47.5		
23	1100	35.8	39.05	.....	.....	35.8	39.05	3.9	4.40	39.7	43.45	+ 3.69
24	1100	42.3		.....	.....	42.3		4.9		47.2		
25	1200	40.0	41.25	.....	.....	40.0	41.25	4.8	4.35	44.8	45.60	+ 5.84
26	1200	42.5		.....	.....	42.5		3.9		46.4		
27	1300	38.4	40.10	.....	.....	39.4	40.10	4.2	4.55	43.6	44.65	+ 4.89
28	1300	40.8		.....	.....	40.8		4.9		45.7		
29	1400	42.8	38.35	.....	.....	42.8	38.35	3.8	4.30	46.6	42.65	+ 2.89
30	1400	33.9		.....	.....	33.9		4.8		38.7		
31	1500	37.2	37.65	.....	.....	37.2	37.65	6.0	4.75	43.2	42.40	+ 2.64
32	1500	38.1		.....	.....	38.1		3.5		41.6		
33	Control	31.5	32.50	.....	.....	31.5	32.50	7.8	7.26	39.3	39.76	
34	Control	31.2		.....	.....	31.2		6.9		38.1		
35	Control	34.8		.....	.....	34.8		7.1		41.9		



TABLE IIb  
CuSO<sub>4</sub> SET—SECOND CROP—GREENHOUSE SOIL

	CuSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	100	9.90	10.25	8.10	9.55	17.0	19.30	5.0	5.60	22.0	24.90	+4.20
2	100	10.60		11.00		21.6		6.2		27.8		
3	200	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
4	200	.....		.....		.....		.....		.....		
5	300	8.15	7.57	11.05	10.02	19.2	17.60	5.5	5.95	24.7	23.55	+2.85
6	300	7.00		9.00		16.0		6.4		22.4		
7	400	14.10	11.10	10.90	10.90	25.0	22.00	7.2	5.85	32.2	27.85	+7.15
8	400	8.10		10.90		19.0		4.5		23.5		
9	500	13.90	10.95	8.10	8.10	22.0	19.00	5.5	7.65	27.7	26.65	+5.95
10	500	8.00		8.00		16.0		9.8		25.8		
11	600	6.10	8.05	12.40	11.20	18.5	19.25	6.0	6.35	24.5	25.60	+4.90
12	600	10.00		10.00		20.0		6.7		26.7		
13	700	13.15	11.75	11.85	12.50	25.0	24.20	6.0	5.10	31.0	29.30	+8.60
14	700	10.25		13.15		23.4		4.2		27.6		
15	800	8.77	10.98	9.23	9.01	18.0	20.00	9.0	7.35	27.0	27.35	+6.65
16	800	13.20		8.80		22.0		5.7		27.7		
17	900	15.45	12.07	9.15	8.22	24.6	20.30	7.3	6.55	31.9	26.85	+6.15
18	900	8.70		7.30		16.0		5.8		21.8		
19	1000	12.45	12.16	15.05	11.39	27.5	23.55	5.5	4.85	33.0	28.40	+7.70
20	1000	11.88		7.72		19.6		4.2		23.8		
21	1100	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
22	1100	.....		.....		.....		.....		.....		
23	1200	7.45	9.27	11.75	13.73	19.2	23.00	5.5	5.00	24.7	28.00	+7.30
24	1200	11.10		15.70		26.8		4.5		31.3		
25	1300	12.75	10.97	8.75	10.02	21.5	21.00	4.9	4.80	26.4	25.80	+5.10
26	1300	9.20		11.30		20.5		4.7		25.2		
27	1400	12.60	12.37	13.20	12.52	25.8	24.90	5.5	5.60	30.3	30.50	+9.80
28	14.00	12.15		11.85		24.0		5.7		29.7		
29	1500	8.45	8.45	10.15	10.15	18.6	18.60	3.2	3.20	21.8	21.80	+1.10
30	1500	.....		.....		.....		.....		.....		
31	Control	6.00	7.13	9.20	9.37	15.2	16.50	4.0	4.20	19.2	20.70	
32	Control	8.26		9.54		17.8		4.4		22.2		

TABLE IIc  
CuSO<sub>4</sub> SET—THIRD CROP—GREENHOUSE SOIL

	CuSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	100	23.90	19.45	2.60	2.80	26.5	22.25	6.5	9.75	33.00	32.00	+5.37
2	100	15.00		3.00		18.0		13.0		31.00		
3*	200	18.25	21.17	5.75	4.32	24.0	25.50	15.5	7.75	31.75	33.25	+6.62
4*	200	24.10		2.90		27.0				34.75		
5	300	19.42	16.71	2.48	2.48	21.8	17.90	10.2	8.35	32.00	26.25	-0.38
6	300	14.00		.....		14.0		6.5		20.50		
7	400	14.05	14.00	4.45	4.25	18.5	18.25	11.0	11.00	29.50	29.25	+2.62
8	400	13.94		4.06		18.0		11.0		29.00		
9	500	16.36	16.03	2.74	3.52	19.0	19.50	9.0	8.25	28.00	27.75	+1.12
10	500	15.70		4.30		20.0		7.5		27.50		
11	600	12.85	17.72	1.15	1.23	14.0	18.95	12.0	10.60	26.00	29.55	+2.92
12	600	22.60		1.30		23.9		9.2		33.10		
13	700	18.05	16.35	3.55	2.85	21.6	19.20	9.0	9.00	30.60	28.20	+1.57
14	700	14.65		2.15		16.8		.....		25.80		
15	800	12.30	15.30	3.70	3.20	16.0	18.50	7.8	6.40	23.80	24.90	-1.73
16	800	18.30		2.70		21.0		5.0		26.00		
17	900	21.20	20.75	.....	4.30	25.5	25.05	7.2	10.60	32.70	35.65	+9.02
18	900	20.30		4.30		24.6		14.0		38.60		
19	1000	17.65	23.15	1.85	2.60	19.5	25.75	10.0	9.50	29.50	35.25	+8.62
20	1000	28.65		3.35		32.0		9.0		41.00		
21	1100	15.20	17.95	1.80	1.50	17.0	19.45	9.0	9.00	26.00	28.45	+1.82
22	1100	20.70		1.20		21.9		9.0		30.90		
23	1200	14.80	15.60	2.70	2.90	17.5	18.50	5.7	6.85	23.20	25.35	-1.28
24	1200	16.40		3.10		19.5		8.0		27.50		
25	1300	18.20	17.95	3.00	2.75	21.2	20.70	4.5	5.25	25.70	25.95	-0.68
26	1300	17.70		2.50		20.2		6.0		26.20		
27	1400	18.90	20.70	3.30	3.30	22.2	22.35	15.3	7.65	29.85	30.00	+3.37
28	1400	22.50		.....		22.5				30.15		
29	1500	14.00	17.65	4.50	4.35	18.5	22.00	11.0	8.05	29.50	30.05	+3.42
30	1500	21.30		4.20		25.5		5.1		30.60		
31	Control	12.40	16.53	2.10	2.25	14.5	18.03	9.8	8.60	24.30	26.63	
32	Control	16.20		2.40		18.6		9.0		27.60		
33	Control	21.00		.....		21.0		7.0		28.00		

\* Second crop.

TABLE IIIa  
CuSO<sub>4</sub>—FIRST CROP—ADOBE SOIL

	CuSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	100	9.50	4.89	.....	3.72	9.5	8.75	0.80	1.10	10.30	9.85	+1.93
2	100	4.28		3.72		8.0		1.40		9.40		
3*	200	4.00	3.50	.....	.....	4.0	3.50	1.20	1.00	5.20	4.50	-2.42
4*	200	3.00		.....		3.0		.80		3.80		
5	300	5.90	4.25	4.10	4.50	10.0	8.75	1.40	1.05	11.40	9.80	+1.88
6	300	2.60		4.90		7.5		.70		8.20		
7	400	2.70	3.43	3.90	4.38	6.5	7.75	1.20	1.55	7.70	9.30	+1.38
8	400	4.15		4.85		9.0		1.90		10.90		
9	500	5.15	6.30	1.95	2.25	7.1	8.65	.85	1.00	7.95	9.65	+1.73
10	500	7.45		2.55		10.2		1.15		11.35		
11	600	5.05	4.53	4.65	4.02	9.7	8.60	1.00	.92	10.70	9.52	+1.60
12	600	4.10		3.40		7.5		.85		8.35		
13	700	3.20	5.18	3.80	3.58	7.0	8.75	.65	.75	7.65	9.50	+1.58
14	700	7.15		3.35		10.5		.85		11.35		
15	800	4.15	4.63	4.35	3.33	8.5	7.95	1.00	.80	9.50	8.75	+0.83
16	800	5.10		2.30		7.4		.60		8.00		
17	900	6.75	7.43	3.05	3.05	9.8	9.00	.85	1.02	10.65	10.00	+2.10
18	900	8.20		.....		8.2		1.20		9.40		
19	1000	2.70	3.10	1.60	1.60	4.3	3.75	.45	.52	4.75	4.42	-3.50
20	1000	3.50		.....		3.5		.60		4.10		
21	1200	4.10	4.95	1.90	1.90	6.0	5.90	.70	.70	6.70	6.60	-1.32
22	1200	5.80		.....		5.8		.70		6.50		
23	1500	2.90	2.90	.60	.60	3.5	3.50	.20	.20	3.70	3.70	-4.22
24†	1500	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	
25†	2000	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	
26	Control	5.20	4.14	2.20	3.06	7.4	7.20	.95	.73	8.35	7.92	
27	Control	3.08		3.92		7.0		.50		7.50		

\* Poor plants due perhaps to location of pots.

† Nos. 24 and 25—no growth.

TABLE IIIb  
CuSO<sub>4</sub> SET—SECOND CROP—ADOBE SOIL

	CuSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	100	6.5	6.25	.....	.....	6.5	6.25	0.45	0.92	6.95	7.18	-1.91
2	100	6.0		.....		6.0		1.40		7.40		
3	200	7.5	7.50	.....	.....	7.5	7.50	1.55	1.52	9.05	9.02	-0.07
4	200	7.5		.....		7.5		1.50		9.00		
5	300	9.0	8.85	.....	.....	9.0	8.85	1.50	1.60	10.50	10.45	+1.36
6	300	8.7		.....		8.7		1.70		10.40		
7	400	6.0	6.25	.....	.....	6.0	6.25	2.55	2.42	8.55	8.68	-0.41
8	400	6.5		.....		6.5		2.30		8.80		
9	500	8.5	8.00	.....	.....	8.5	8.00	1.95	1.95	10.45	9.95	+0.86
10	500	7.5		.....		7.5		1.95		9.45		
11	600	8.8	9.40	.....	.....	8.8	9.40	3.60	2.95	12.40	12.35	+3.26
12	600	10.0		.....		10.0		2.30		12.30		
13	700	10.2	9.35	.....	.....	10.2	9.35	2.10	1.95	12.30	11.30	+2.21
14	700	8.5		.....		8.5		1.80		10.30		
15	800	10.5	9.85	.....	.....	10.5	9.85	1.40	1.60	11.90	11.45	+2.36
16	800	9.2		.....		9.2		1.80		11.00		
17	900	9.5	10.60	.....	.....	9.5	10.60	2.00	2.00	11.50	12.60	+3.51
18	900	11.7		.....		11.7		2.00		13.70		
19	1000	8.8	7.90	.....	.....	8.8	7.90	2.00	2.75	10.80	10.65	+1.56
20	1000	7.0		.....		7.0		3.50		10.50		
21	1200	7.2	7.25	.....	.....	7.2	7.25	1.65	1.50	8.85	8.75	-0.34
22	1200	7.3		.....		7.3		1.35		8.65		
23	1500	10.0	9.00	.....	.....	10.0	9.00	1.30	1.32	11.30	10.32	+1.23
24	1500	8.0		.....		8.0		1.35		9.35		
25	2000	5.1	5.10	.....	.....	5.1	5.10	0.32	.032	5.42	5.42	-3.67
26	Control	7.0	7.25	.....	.....	7.0	7.25	1.93	1.84	8.93	9.09	
27	Control	7.5		.....		7.5		1.75		9.25		

TABLE IV  
CuSO<sub>4</sub>—FIRST CROP—OAKLEY SOIL

	CuSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	100	4.78	4.52	1.72	1.79	6.5	6.30	2.10	1.85	8.60	8.15	+2.15
2	100	4.25		1.85		6.1		1.60		7.70		
3	200	3.68	3.43	2.32	2.52	6.0	5.95	2.70	2.40	8.70	8.35	+2.35
4	200	3.17		2.73		5.9		2.10		8.00		
5	300	3.90	3.80	2.30	2.40	6.2	6.20	1.30	1.10	7.30	7.20	+1.20
6	300	3.70		2.50		6.2		.90		7.10		
7	400	2.40	2.40	1.60	1.60	4.0	4.00	.55	.55	4.55	4.55	—1.45
8	400	.....		.....		....		.....		.....		
9	500	2.90	2.75	1.20	1.30	4.1	4.05	.15	.15	4.25	4.20	—1.80
10	500	2.60		1.40		4.0		.15		4.15		
11	600	2.85	3.53	2.35	2.67	5.2	6.20	.....	.....	5.20	6.20	+0.20
12	600	4.20		3.00		7.2		.....		7.20		
13	700	2.40	2.80	1.30	1.40	3.7	4.20	.....	.....	3.70	4.20	—1.80
14	700	3.20		1.50		4.7		.....		4.70		
15	800	1.20	1.20	.....	.....	1.2	1.20	.10	.10	1.30	1.30	—4.70
16	800	.....		.....		....		.....		.....		
17	900	.20	.20	.....	.....	.2	.20	.....	.....	.20	.20	—5.80
18	900	.....		.....		....		.....		.....		
19	1000	.....	.....	.....	.....	....	.....	.....	.....	.....	.....	.....
20	1000	.....		.....		....		.....		.....		
21	1200	.....	.....	.....	.....	....	.....	.....	.....	.....	.....	.....
22	1200	.....		.....		....		.....		.....		
23	Control	2.80	3.40	1.30	1.30	4.1	4.05	1.60	1.95	5.70	6.00	
24	Control	4.00		.....		4.0		2.30		6.30		

TABLE Va  
ZnSO<sub>4</sub> SET—FIRST CROP—GREENHOUSE SOIL

	ZnSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	100	38.9	37.45	.....	.....	38.9	37.45	8.6	8.55	47.5	46.00	+ 4.30
2	100	36.0	.....	.....	.....	36.0	.....	8.5	.....	44.5	.....	.....
3	300	38.9	36.55	.....	.....	38.9	36.55	7.5	7.60	46.4	44.15	+ 2.45
4	300	34.2	.....	.....	.....	34.2	.....	7.7	.....	41.9	.....	.....
5	500	37.4	39.10	.....	.....	37.4	39.10	6.5	9.90	43.9	49.00	+ 7.30
6	500	40.8	.....	.....	.....	40.8	.....	13.3	.....	54.1	.....	.....
7	700	46.8	40.30	.....	.....	46.8	40.30	8.6	10.60	55.4	50.90	+ 9.20
8	700	33.8	.....	.....	.....	33.8	.....	12.6	.....	46.4	.....	.....
9	900	41.4	41.30	.....	.....	41.4	41.30	8.1	8.00	49.5	49.30	+ 7.60
10	900	41.2	.....	.....	.....	41.2	.....	7.9	.....	49.1	.....	.....
11	1100	41.1	42.30	.....	.....	41.1	42.30	7.7	9.25	48.8	51.55	+ 9.85
12	1100	43.5	.....	.....	.....	43.5	.....	10.8	.....	54.3	.....	.....
13	1200	42.2	41.40	.....	.....	42.2	41.40	8.3	8.50	50.5	49.90	+ 8.20
14	1200	40.6	.....	.....	.....	40.6	.....	8.7	.....	49.3	.....	.....
15	1300	31.2	31.35	.....	.....	31.2	31.35	12.0	12.00	43.2	43.35	+ 1.65
16	1300	31.5	.....	.....	.....	31.5	.....	.....	.....	43.5	.....	.....
17	1400	36.7	34.85	.....	.....	36.7	34.85	10.8	11.15	47.5	46.00	+ 4.30
18	1400	33.0	.....	.....	.....	33.0	.....	11.5	.....	44.5	.....	.....
19	1500	38.5	38.10	.....	.....	38.5	38.10	6.2	7.50	44.7	45.60	+ 3.90
20	1500	37.7	.....	.....	.....	37.7	.....	8.8	.....	46.5	.....	.....
21	1600	37.0	36.00	.....	.....	37.0	36.00	11.3	12.15	48.3	48.15	+ 6.45
22	1600	35.0	.....	.....	.....	35.0	.....	13.0	.....	48.0	.....	.....
23	1700	35.8	36.80	.....	.....	35.8	36.80	9.3	10.60	45.1	47.40	+ 5.70
24	1700	37.8	.....	.....	.....	37.8	.....	11.9	.....	49.7	.....	.....
25	1800	38.7	38.10	.....	.....	38.7	38.10	10.0	10.20	48.7	48.30	+ 6.60
26	1800	37.5	.....	.....	.....	37.5	.....	10.4	.....	47.9	.....	.....
27	1900	43.8	41.90	.....	.....	43.8	41.90	13.0	13.15	56.8	55.05	+13.35
28	1900	40.0	.....	.....	.....	40.0	.....	13.3	.....	53.3	.....	.....
29	2000	31.0	35.60	.....	.....	31.0	35.60	10.4	10.85	41.4	46.45	+ 4.75
30	2000	40.2	.....	.....	.....	40.2	.....	11.3	.....	51.5	.....	.....
31	Control	30.2	33.40	.....	.....	30.2	33.40	8.5	8.30	38.7	.....	.....
32	Control	36.6	.....	.....	.....	36.6	.....	8.1	.....	44.7	41.70	.....

TABLE Vb  
ZnSO<sub>4</sub> SET—SECOND CROP—GREENHOUSE SOIL

	ZnSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	100	12.50	12.40	3.50	3.50	16.00	15.90	3.5	3.55	19.50	19.45	+2.95
2	100	12.30		3.50		15.80		3.6		19.40		
3	300	8.60	7.95	4.40	4.05	13.00	12.00	6.0	5.80	19.00	17.80	+1.30
4	300	7.30		3.70		11.00		5.6		16.60		
5	500	4.82	4.65	4.38	5.05	9.20	9.70	6.3	4.65	15.50	14.35	—2.15
6	500	4.48		5.72		10.20		3.0		13.20		
7	700	8.80	7.84	.....	6.11	8.80	10.90	4.6	4.30	13.40	15.20	—1.30
8	700	6.89		6.11		13.00		4.0		17.00		
9	900	5.80	5.77	6.70	4.98	12.50	10.75	3.0	4.10	15.50	14.85	—1.65
10	900	5.74		3.26		9.00		5.2		14.20		
11	1100	4.93	7.96	3.07	3.07	8.00	9.50	4.3	4.40	12.30	13.90	—2.60
12	1100	11.00		.....		11.00		4.5		15.50		
13	1200	7.65	7.40	.....	6.35	14.00	13.75	3.7	3.50	17.70	17.25	—0.75
14	1200	7.15		6.35		13.50		3.3		16.80		
15	1300	6.20	7.55	4.30	4.44	10.50	12.00	3.8	3.90	14.30	15.90	—0.60
16	1300	8.91		4.59		13.50		4.0		17.50		
17	1400	6.29	7.66	4.71	4.74	11.00	12.40	2.8	2.50	13.80	14.90	—1.60
18	1400	9.03		4.77		13.80		2.2		16.00		
19	1500	10.40	7.92	.....	4.55	10.40	10.20	2.5	3.00	12.90	13.20	—3.30
20	1500	5.45		4.55		10.00		3.5		13.50		
21	1600	5.08	5.49	5.72	4.41	10.80	9.90	3.7	3.35	14.50	13.25	—3.25
22	1600	5.90		3.10		9.00		3.0		12.00		
23	1700	5.08	4.41	3.42	4.24	8.50	8.65	3.2	3.05	11.70	11.70	—4.80
24	1700	3.74		5.06		8.80		2.9		11.70		
25	1800	6.45	6.77	4.05	3.72	10.50	10.50	2.9	3.20	13.40	13.70	—2.80
26	1800	7.10		3.40		10.50		3.5		14.00		
27	1900	6.36	5.97	3.04	2.63	9.40	8.10	1.8	2.80	11.20	10.90	—5.60
28	1900	4.58		2.22		6.80		3.8		10.60		
29	2000	6.52	6.17	3.46	3.30	10.00	9.50	2.5	2.25	12.50	11.75	—4.85
30	2000	5.85		3.15		9.00		2.0		11.00		
31	Control	7.50	8.28	4.70	5.42	12.20	13.70	2.6	2.80	14.80	16.50	
32	Control	9.06		6.14		15.20		3.0		17.20		

TABLE Vc  
 $\text{ZnSO}_4$  SET—THIRD CROP—GREENHOUSE SOIL

	$\text{ZnSO}_4$ added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1*	100	23.74	21.32	8.62	5.36	32.29	26.65	9.5	9.00	41.7	35.65	+ 4.89
2	100	18.90		2.10		21.00		8.5		29.5		
3	300	19.50	19.70	2.50	2.15	21.90	21.80	5.0	5.65	26.9	27.45	— 3.31
4	300	19.90		1.80		21.70		6.3		28.0		
5	500	20.70	17.12	3.80	2.87	24.50	20.00	5.0	4.85	29.5	24.85	— 5.91
6	500	13.55		1.95		15.50		4.7		20.2		
7	700	18.32	17.99	2.68	2.52	21.00	20.50	4.8	4.75	25.8	25.25	— 5.51
8	700	17.67		2.37		20.00		4.7		24.7		
9	900	17.40	17.05	1.60	2.45	19.00	19.50	5.5	5.05	24.5	24.55	— 6.21
10	900	16.70		3.30		20.00		4.6		24.6		
11	1100	18.60	17.91	1.80	2.29	20.40	20.20	5.7	5.10	26.1	25.30	— 5.46
12	1100	17.22		2.78		20.00		4.5		24.5		
13	1200	21.75	20.82	2.25	1.62	24.00	22.50	5.0	4.60	29.0	27.10	— 3.66
14	1200	19.90		1.10		21.00		4.2		25.2		
15	1300	15.75	13.65	.75	1.60	16.50	15.25	4.5	4.50	21.0	19.75	—11.01
16	1300	11.55		2.45		14.00		4.3		18.5		
17	1400	15.15	16.55	.85	.75	16.00	17.30	4.0	4.00	20.0	21.30	— 9.46
18	1400	17.95		.65		18.60		....		22.6		
19	1500	14.35	12.90	3.15	2.10	17.40	15.00	5.5	4.25	22.9	19.25	—11.51
20	1500	11.55		1.05		12.60		3.0		15.6		
21	1600	14.55	13.72	2.55	2.40	17.10	16.15	5.5	5.15	22.6	21.30	— 9.46
22	1600	12.90		2.30		15.20		4.8		20.0		
23	1700	9.98	13.45	1.02	1.52	11.00	15.00	3.5	4.00	15.0	19.00	—11.68
24	1700	16.93		2.03		19.00		4.5		23.0		
25	1800	12.75	13.00	1.25	1.25	14.00	14.25	3.0	2.70	17.0	16.95	—13.73
26	1800	13.25		1.25		14.50		2.4		16.9		
27	1900	13.80	13.34	2.50	2.06	16.30	15.40	2.4	2.35	18.7	17.75	—13.01
28	1900	12.88		1.62		14.50		2.3		16.8		
29	2000	11.56	11.17	2.44	1.67	14.00	21.85	2.0	2.65	16.0	15.75	—15.01
30	2000	10.79		0.91		11.70		3.3		15.5		
31	Control	16.50	20.03	2.10	2.25	18.60	21.53	10.5	9.23	29.1	30.76	
32	Control	22.10		2.40		24.50		8.2		32.7		
33	Control	21.50		.....		21.50		9.0		30.5		

\* Contaminated by rain.



TABLE VIa  
 FeSO<sub>4</sub> SET—FIRST CROP—GREENHOUSE SOIL

	FeSO <sub>4</sub> added to soil in terms of percentage	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	0.1	40.0	39.95	.....	.....	40.0	39.95	10.6	9.05	50.6	49.00	+8.20
2	0.1	39.9		.....		39.9		7.5		47.4		
3	0.2	39.7	35.45	.....	.....	39.7	35.45	6.0	7.20	45.7	42.65	+1.85
4	0.2	31.2		.....		31.2		8.4		39.6		
5	0.3	43.7	40.30	.....	.....	43.7	40.30	7.0	9.45	50.0	49.75	+8.95
6	0.3	36.9		.....		36.9		11.9		48.8		
7	0.4	41.0	39.90	.....	.....	41.0	39.90	6.3	8.65	47.3	48.55	+7.85
8	0.4	38.8		.....		38.8		11.0		49.8		
9	0.5	34.0	35.25	.....	.....	34.0	35.25	9.2	8.40	43.2	43.65	+2.85
10	0.5	36.5		.....		36.5		7.6		44.1		
11	0.6	38.5	39.10	.....	.....	38.5	39.10	9.7	8.50	48.2	47.60	+6.80
12	0.6	39.7		.....		39.7		7.3		47.0		
13	0.7	28.2	36.30	.....	.....	28.2	36.30	13.4	9.60	41.6	45.90	+5.10
14	0.7	44.4		.....		44.4		5.8		50.2		
15	0.8	32.8	38.20	.....	.....	32.8	38.20	4.2	5.65	37.0	43.85	+3.05
16	0.8	43.6		.....		43.6		7.1		50.7		
17	0.9	37.1	36.80	.....	.....	37.1	36.80	6.9	6.70	44.0	43.50	+2.70
18	0.9	36.5		.....		36.5		6.5		43.0		
19	1.0	38.7	34.85	.....	.....	28.7	34.85	5.9	6.35	34.6	41.20	+0.40
20	1.0	41.0		.....		41.0		6.8		47.8		
21	Control	34.8	32.50	.....	.....	34.8	32.50	8.5	8.30	43.3	40.80	
22	Control	30.2		.....		30.2		8.1		38.3		

TABLE VIb  
 FES<sub>2</sub>O<sub>3</sub> SET—SECOND CROP—GREENHOUSE SOIL

	FeSO <sub>4</sub> added to soil in terms of percentage	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	0.1	11.20	8.83	2.60	5.16	13.80	14.00	4.3	4.90	18.10	18.90	+2.17
2	0.1	6.47		7.73		14.20		5.5		19.70		
3	0.2	9.90	8.32	6.50	6.47	16.40	14.80	4.7	4.95	21.10	19.75	+3.02
4	0.2	6.75		6.45		13.20		5.2		18.40		
5	0.3	9.30	7.40	6.50	5.50	15.80	12.90	4.7	5.35	20.50	18.25	+1.52
6	0.3	5.50		4.50		10.00		6.0		16.00		
7	0.4	9.45	8.13	4.55	5.01	14.00	13.15	3.0	3.10	17.00	16.25	—0.48
8	0.4	6.82		5.48		12.30		3.2		15.50		
9	0.5	.....		.....		.....		.....		.....		
10	0.5	8.25	8.25	6.75	6.75	15.00	15.00	3.5	3.50	18.50	18.50	+1.77
11	0.6	6.90	7.49	4.30	4.16	11.20	11.65	3.5	3.60	14.70	15.25	—1.48
12	0.6	8.08		4.02		12.10		3.7		15.80		
13	0.7	6.27	6.33	5.03	6.06	11.30	12.40	3.0	3.00	14.30	15.40	—1.33
14	0.7	6.40		7.10		13.50		3.0		16.50		
15	0.8	8.50	7.20	6.50	4.05	15.00	11.20	2.7	2.40	17.70	13.60	—3.13
16	0.8	5.90		1.60		7.50		2.1		9.60		
17	0.9	.....		.....		.....		.....		.....		
18	0.9	6.80		.....		6.80	6.80	2.5	2.50	9.30	9.30	—7.43
19	1.0	5.40	5.40	1.10	1.10	6.50	6.50	2.5	2.50	9.00	9.00	—7.73
20	1.0	.....		.....		.....		.....		.....		
21	Control	7.30	7.68	4.70	5.42	12.00	13.13	2.0	3.60	14.00		
22	Control	8.06		6.14		15.20		4.4		19.60	16.73	
23	Control	12.20		.....		12.20		4.4		16.60		

TABLE VIc  
 FESO<sub>4</sub> SET—THIRD CROP—GREENHOUSE SOIL

	FeSO <sub>4</sub> added to soil in terms of percentage	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	0.1	19.00	17.55	.....	0.47	19.0	19.90	7.0	7.50	26.0	27.40	— 3.15
2	0.1	16.10		4.70		20.8		8.0		28.8		
3	0.2	20.05	17.80	1.85	3.05	21.9	20.85	8.2	8.10	29.9	28.95	— 1.60
4	0.2	15.55		4.25		19.8		8.0		28.0		
5	0.3	13.75	14.27	5.65	4.82	19.4	19.60	4.4	4.80	23.8	24.40	— 6.15
6	0.3	15.80		4.00		19.8		5.2		25.0		
7	0.4	20.70	19.17	4.80	4.07	25.5	23.75	5.8	6.40	31.3	30.15	— 0.40
8	0.4	18.65		3.35		22.0		7.0		29.0		
9*	0.5	34.80	25.87	.....	4.05	34.8	27.90	4.0	6.75	38.8	34.65	+ 4.10
10	0.5	16.95		4.05		21.0		9.5		30.5		
11	0.6	14.45	13.87	4.55	4.12	19.0	18.00	5.0	5.40	24.0	23.40	— 7.15
12	0.6	13.30		3.70		17.0		5.8		22.8		
13	0.7	25.62	23.31	4.40	4.10	30.2	27.50	6.2	8.75	36.4	36.35	+ 5.85
14	0.7	21.00		3.80		24.8		11.5		36.3		
15	0.8	31.90	27.41	2.60	3.23	34.5	30.65	6.0	8.15	40.5	38.80	+ 8.25
16	0.8	22.93		3.87		26.8		10.3		37.1		
17	0.9	28.90	21.52	2.30	4.57	31.2	26.10	6.7	7.00	37.9	33.10	+ 2.55
18	0.9	14.15		6.85		21.0		7.3		28.3		
19	1.0	20.10	27.60	8.90	7.40	29.0	35.00	9.7	8.95	38.7	43.95	+13.40
20*	1.0	35.10		5.90		41.0		8.2		49.2		
21	Control	22.40	19.30	2.10	2.25	24.5	21.55	7.5	9.00	32.0	30.55	
22	Control	16.20		2.40		18.6		10.5		29.1		

\* Failed to grow second crop.

TABLE VIIa  
 P<sub>2</sub>SO<sub>5</sub> SET—FIRST CROP—GREENHOUSE SOIL

	P <sub>2</sub> SO <sub>5</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	200	16.5	21.00	.....	.....	16.5	21.00	4.4	4.40	20.9	25.40	—14.95
2	200	25.5	.....	.....	.....	25.5	.....	4.4	.....	29.9	.....	.....
3	300	19.5	21.30	.....	.....	19.5	21.30	5.1	4.05	24.6	25.35	—15.00
4	300	23.1	.....	.....	.....	23.1	.....	3.0	.....	26.1	.....	.....
5	400	21.3	22.20	.....	.....	21.3	22.20	5.2	4.75	26.5	26.95	—13.40
6	400	23.1	.....	.....	.....	23.1	.....	4.3	.....	27.4	.....	.....
7*	500	18.4	18.40	.....	.....	18.4	18.40	3.8	3.80	22.2	22.20	—18.15
8	500	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
9	600	22.2	23.30	.....	.....	22.2	23.30	3.2	2.80	25.4	26.10	—14.25
10	600	24.4	.....	.....	.....	24.4	.....	2.4	.....	26.8	.....	.....
11	700	24.0	23.60	.....	.....	24.0	23.60	4.0	3.00	28.0	26.60	—13.75
12	700	23.2	.....	.....	.....	23.2	.....	2.0	.....	25.2	.....	.....
13	800	26.8	21.90	.....	.....	26.8	21.90	6.8	4.40	33.6	26.30	—14.05
14	800	17.0	.....	.....	.....	17.0	.....	2.0	.....	19.0	.....	.....
15	900	26.8	28.55	.....	.....	26.8	28.55	3.8	4.20	30.6	32.75	— 7.60
16	900	30.3	.....	.....	.....	30.3	.....	4.6	.....	34.9	.....	.....
17	1000	22.0	21.75	.....	.....	22.0	21.75	5.4	3.95	27.4	25.70	—14.65
18	1000	21.5	.....	.....	.....	21.5	.....	2.5	.....	24.0	.....	.....
18	1100	18.2	23.35	.....	.....	18.2	23.35	5.3	5.40	23.5	28.75	—11.60
20	1100	28.5	.....	.....	.....	28.5	.....	5.5	.....	34.0	.....	.....
21	1200	21.9	23.95	.....	.....	21.9	23.95	3.6	4.55	25.5	28.50	—11.85
22	1200	26.0	.....	.....	.....	26.0	.....	5.5	.....	31.5	.....	.....
23	1300	23.8	25.85	.....	.....	23.8	25.85	6.8	4.40	30.6	30.25	—10.10
24	1300	27.9	.....	.....	.....	27.9	.....	2.0	.....	29.9	.....	.....
25	1400	25.0	21.60	.....	.....	25.0	21.60	2.9	3.55	27.9	25.15	—15.20
26	1400	18.2	.....	.....	.....	18.2	.....	4.2	.....	22.4	.....	.....
27	1500	17.8	22.25	.....	.....	17.8	22.25	1.8	2.50	19.6	24.75	—15.60
28	1500	26.7	.....	.....	.....	26.7	.....	3.2	.....	29.9	.....	.....
29	Control	34.8	30.00	.....	.....	34.8	30.00	6.5	10.35	41.3	40.35	.....
30	Control	25.2	.....	.....	.....	25.2	.....	14.2	.....	39.4	.....	.....

\* Poor plants.

TABLE VIIIb

PbSO<sub>4</sub> SET—SECOND CROP—GREENHOUSE SOIL

	PbSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1*	200	8.0	7.00	.....	.....	8.0	7.00	2.0	2.50	10.0	9.50	—8.70
2*	200	6.0		.....		6.0		3.0		9.0		
3	300	19.5	20.25	.....	.....	19.5	20.25	6.2	4.85	25.7	25.10	+6.90
4	300	21.0		.....		21.0		3.5		24.5		
5*	400	6.5	8.00	.....	.....	6.5	8.00	2.5	2.25	9.0	10.25	—9.95
6*	400	9.5		.....		9.5		2.0		11.5		
7	500	22.0	17.00	.....	.....	22.0	17.00	3.0	3.90	25.0	20.90	+2.70
8*	500	12.0		.....		12.0		4.8		16.8		
9	600	22.6	21.50	.....	.....	22.6	21.50	3.1	3.65	25.7	24.70	+6.50
10	600	19.5		.....		19.5		4.2		23.7		
11*	700	11.0	13.70	.....	.....	11.0	13.70	2.1	2.80	13.1	16.50	—1.70
12	700	16.4		.....		16.4		3.5		19.9		
13	800	16.2	14.60	.....	.....	16.2	14.60	3.2	2.95	19.4	17.55	—1.65
14	800	13.0		.....		13.0		2.7		15.7		
15	900	14.2	14.10	.....	.....	14.2	14.10	4.0	3.00	18.2	17.10	—1.10
16	900	14.0		.....		14.0		2.0		16.0		
17	1000	15.4	16.20	.....	.....	15.4	16.20	2.9	2.70	18.3	18.90	+0.70
18	1000	17.0		.....		17.0		2.5		19.5		
19	1100	11.0	10.75	.....	.....	11.0	10.75	2.7	2.00	13.7	12.75	+5.45
20	1100	10.5		.....		10.5		1.3		11.8		
21	1200	19.8	18.20	.....	.....	19.8	18.20	2.4	2.20	22.2	20.40	+2.20
22	1200	16.6		.....		16.6		2.0		18.6		
23	1300	14.8	15.80	.....	.....	14.8	15.80	2.7	2.45	17.5	18.25	+0.05
24	1300	16.8		.....		16.8		2.2		19.0		
25	1400	21.0	19.00	.....	.....	21.0	19.00	3.0	4.00	24.0	23.00	+4.80
26	1400	17.0		.....		17.0		5.0		22.0		
27	1500	16.4	14.90	.....	.....	16.4	14.90	2.2	2.65	18.6	17.55	—0.65
28	1500	13.4		.....		13.4		3.1		16.5		
29	Control	15.2	14.00	.....	.....	15.2	14.00	4.4	4.20	19.6	18.20	
30	Control	12.8		.....		12.8		4.0		16.8		

\* Plants partly damaged by mice.

TABLE VIIc  
PbSO<sub>4</sub> SET—THIRD CROP—GREENHOUSE SOIL

	PbSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	200	16.20	15.10	2.30	2.30	18.5	16.25	8.0	8.00	26.5	24.25	— 4.32
2	200	14.00		.....		1.4		8.0		22.0		
3	300	15.70	17.45	2.30	2.30	18.0	19.75	7.2	7.15	25.2	26.85	— 1.67
4	300	19.20		2.30		21.5		7.1		28.6		
5*	400	9.80	11.70	2.40	2.15	12.2	13.85	4.2	3.80	16.4	17.65	—10.92
6*	400	13.60		1.90		15.5		3.4		18.9		
7	500	24.30	24.35	1.90	2.15	26.2	26.50	6.9	7.95	33.1	34.45	+ 5.88
8	500	24.40		2.40		26.8		7.0		33.8		
9	600	24.70	24.60	.....	.....	24.7	24.60	5.7	7.75	30.4	32.35	+ 3.78
10	600	24.50		.....		24.5		9.8		34.3		
11	700	20.90	20.25	4.00	4.00	24.9	22.75	9.5	9.15	34.4	31.90	+ 3.33
12	700	19.60		.....		19.6		8.8		28.4		
13	800	22.35	21.17	2.65	2.65	25.0	22.70	5.9	6.10	30.9	28.60	+ 0.03
14	800	20.00		.....		20.0		6.3		26.3		
15	900	18.80	20.67	5.60	4.27	24.4	24.95	5.8	7.80	32.2	33.75	+ 5.18
16	900	22.55		2.95		25.5		9.8		35.3		
17	1000	19.72	20.63	2.48	2.82	22.2	23.45	7.0	7.00	29.2	30.45	+ 1.88
18	1000	21.55		3.15		24.7		7.0		31.7		
19	1100	29.22	25.37	4.67	3.06	33.9	28.45	9.0	6.60	40.5	35.05	+ 6.48
20	1100	21.55		1.45		23.0		4.2		29.6		
21	1200	21.05	21.07	3.25	3.22	24.3	24.30	7.0	7.25	31.3	31.55	+ 2.98
22	1200	21.10		3.20		24.3		7.5		31.8		
23	1300	20.77	18.76	5.03	3.14	25.8	21.90	6.8	5.90	31.6	27.80	— 0.77
24	1300	16.75		1.25		18.0		6.0		24.0		
25	1400	17.00	17.45	5.00	4.40	22.0	21.85	7.0	8.00	29.0	29.85	+ 1.28
26	1400	17.90		3.80		21.7		9.0		30.7		
27	1500	16.20	18.90	1.50	1.50	17.7	19.65	4.2	6.10	23.8	25.75	— 2.82
28	1500	21.60		.....		21.6		8.0		29.6		
29	Control	16.50	18.65	2.10	2.25	18.6	19.77	10.5	8.80	29.1	28.57	
30	Control	22.10		2.40		24.5		8.2		32.7		
31	Control	14.50		.....		14.5		7.5		22.0		
32	Control	21.50		.....		21.5		9.0		30.5		

\* Plants partly damaged by mice.

TABLE VIIIa  
 POTASH ALUM SET—FIRST CROP—GREENHOUSE SOIL

	Potash Alum added at rate of K <sub>2</sub> O per acre	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	300	38.8	39.20	.....	.....	38.8	39.20	6.6	9.15	45.4	48.35	+8.59
2	300	39.6		.....		39.6		11.7		51.3		
3	400	38.4	38.30	.....	.....	38.4	38.30	8.5	7.80	46.9	46.10	+6.34
4	400	38.2		.....		38.2		7.1		45.3		
5	500	29.6	35.90	.....	.....	29.6	35.90	7.7	7.00	37.3	42.90	+3.14
6	500	42.4		.....		42.4		6.3		48.5		
7	600	38.8	39.50	.....	.....	38.8	39.50	7.5	7.85	46.3	47.35	+7.59
8	600	40.2		.....		40.2		8.2		48.4		
9	700	36.3	36.50	.....	.....	36.3	36.50	5.5	7.15	41.8	43.65	+3.89
10	700	36.7		.....		36.7		8.8		45.5		
11	800	34.4	33.45	.....	.....	34.4	33.45	5.7	5.70	40.1	39.15	—0.61
12	800	32.5		.....		32.5		....		38.2		
13	900	36.8	39.40	.....	.....	36.8	39.40	6.8	6.85	43.6	46.25	+6.49
14	900	42.0		.....		42.0		6.9		48.9		
15	1000	34.2	36.80	.....	.....	34.2	36.80	8.0	7.65	42.2	44.45	+4.69
16	1000	39.4		.....		39.4		7.3		46.7		
17	2000	35.5	34.95	.....	.....	35.5	34.95	6.1	5.20	41.6	40.15	+0.39
18	2000	34.4		.....		34.4		4.3		38.7		
19	Control	31.5	32.50	.....	.....	31.5	32.50	7.8	7.26	39.3	39.76	
20	Control	31.2		.....		31.2		6.9		38.1		
21	Control	34.8		.....		34.8		7.1		41.9		

TABLE VIIIb  
 POTASH ALUM SET—SECOND CROP—GREENHOUSE SOIL

	Potash Alum added at rate of K <sub>2</sub> O per acre	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1*	300	17.88	13.99	15.12	11.81	33.1	25.80	7.8	6.40	40.8	32.20	+11.61
2	300	10.10		8.50		18.6		5.0		23.6		
3	400	8.70	8.90	11.80	11.75	20.5	20.70	6.0	6.60	26.5	27.15	+ 6.56
4	400	9.10		11.70		20.8		7.2		28.0		
5	500	13.70	11.75	8.80	9.40	22.5	21.15	5.0	6.60	27.5	27.15	+ 6.56
6	500	9.80		10.00		19.8		7.0		26.8		
7	600	13.20	11.35	8.80	9.15	22.0	20.50	7.0	6.75	29.0	27.25	+ 6.66
8	600	9.50		9.50		19.0		6.5		25.5		
9	700	9.50	10.82	7.70	8.17	17.2	19.00	7.0	7.00	24.2	26.00	+ 5.41
10	700	12.15		8.65		20.8		7.0		27.8		
11	800	15.90	11.85	11.10	9.85	27.0	21.70	6.8	7.65	33.8	29.35	+ 8.76
12	800	7.80		8.60		16.4		8.5		24.9		
13	900	9.65	9.85	10.85	10.90	20.5	20.75	5.8	5.80	26.3	26.50	+ 5.91
14	900	10.05		10.95		21.0		5.8		26.8		
15	1000	13.30	12.00	10.50	9.50	23.8	21.50	7.5	7.50	31.3	29.00	+ 8.41
16	1000	10.70		8.50		19.2		7.5		26.7		
17	2000	11.00	10.60	11.60	10.90	22.6	21.50	4.0	4.70	26.6	26.20	+ 5.61
18	2000	10.20		10.20		20.4		5.4		25.8		
19	Control	5.66	7.05	9.54	9.29	15.2	16.33	4.0	4.26	19.2	20.59	
20	Control	8.48		9.32		17.8		4.4		22.2		
21	Control	7.00		9.00		16.0		4.4		20.4		

\* Contaminated by rain water.



TABLE VIIIc  
 POTASH ALUM SET—THIRD CROP—GREENHOUSE SOIL

	Potash Alum added at rate of K <sub>2</sub> O per acre	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	300	14.03	15.51	.....	2.78	14.3	16.90	8.2	7.80	22.5	24.70	—2.73
2.	300	16.72		2.78		19.5		7.4		26.9		
3	400	12.65	12.70	1.85	2.05	14.5	14.75	7.3	6.65	21.8	21.40	—6.03
4	400	12.75		2.25		15.0		6.0		21.0		
5	500	11.88	11.29	1.32	2.31	13.2	13.60	5.2	4.85	18.4	18.45	—8.98
6	500	10.70		3.30		14.0		4.5		18.5		
7	600	12.44	14.64	1.76	2.20	14.2	16.85	6.0	5.50	19.7	22.35	—5.08
8	600	16.85		2.65		19.5		5.0		24.5		
9	700	12.00	11.25	.....	.....	12.0	11.25	5.7	6.60	17.7	17.85	—9.58
10	700	10.50		.....		10.5		7.5		18.0		
11	800	15.50	14.62	.....	1.45	15.5	15.35	7.5	7.50	23.0	22.85	—4.58
12	800	13.75		1.45		15.2		7.5		22.7		
13	900	11.70	11.54	1.20	2.64	12.9	14.15	6.7	6.45	19.6	20.60	—6.83
14	900	11.38		4.08		15.4		6.2		21.6		
15	1000	12.50	12.67	2.10	1.92	14.6	14.60	5.2	4.60	19.8	19.20	—8.23
16	1000	21.85		1.75		14.6		4.0		18.6		
17	2000	13.60	15.32	2.40	2.62	16.0	18.00	7.4	6.50	23.4	24.50	—2.93
18	2000	17.05		2.95		20.0		5.6		25.6		
19	Control	19.40	16.70	2.10	2.25	21.5	18.20	8.2	9.23	29.7	27.43	
20	Control	16.20		2.40		18.6		9.0		27.6		
21	Control	14.50		.....		14.5		10.5		25.0		

TABLE IXa  
MNSO<sub>4</sub> SET—FIRST CROP—GREENHOUSE SOIL

	MnSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	500	47.5	51.00	.....	.....	47.5	51.00	11.5	12.00	59.0	63.00	+10.87
2	500	54.5		.....		54.5		12.5		67.0		
3	1000	54.3	54.15	.....	.....	54.3	54.15	11.5	12.25	65.8	66.40	+14.27
4	1000	54.0		.....		54.0		13.0		67.0		
5	1500	56.8	61.90	.....	.....	56.8	61.90	13.5	12.50	70.3	74.40	+22.27
6	1500	67.0		.....		67.0		11.5		78.5		
7	2000	45.6	44.40	.....	.....	45.6	44.40	7.6	8.30	53.2	52.70	+ 0.57
8	2000	43.2		.....		43.2		9.0		52.2		
9	2500	49.2	46.70	.....	.....	49.2	46.70	9.5	9.60	56.2	55.05	+ 2.92
10	2500	44.2		.....		44.2		9.7		53.9		
11	3000	42.0	44.75	.....	.....	42.0	44.75	10.5	9.00	52.5	53.75	+ 1.62
12	3000	47.5		.....		47.5		7.5		55.0		
13	3500	45.7	40.75	.....	.....	45.7	40.75	9.2	9.45	54.9	50.20	— 1.93
14	3500	35.8		.....		35.8		9.7		45.5		
15	4000	39.0	41.75	.....	.....	39.0	41.75	8.0	7.75	47.0	49.50	— 2.63
16	4000	44.5		.....		44.5		7.5		52.0		
17	4500	43.0	43.00	.....	.....	43.0	43.00	10.7	9.85	53.7	52.85	+ 0.72
18	4500	43.0		.....		43.0		9.0		52.0		
19	5000	42.0	45.50	.....	.....	42.0	45.50	8.0	7.50	50.0	53.00	+ 0.87
20	5000	49.0		.....		49.0		7.0		56.0		
21	5500	42.8	39.10	.....	.....	42.8	39.10	6.5	6.80	49.3	45.90	— 6.23
22	5500	35.4		.....		35.4		7.1		42.5		
23	6000	41.0	45.85	.....	.....	41.0	45.85	8.0	7.35	49.0	53.20	+ 1.07
24	6000	50.7		.....		50.7		6.7		57.4		
25	Control	41.1	41.46	.....	.....	41.4	41.46	12.5	10.66	53.9	52.13	
26	Control	43.0		.....		43.0		11.0		54.0		
27	Control	40.0		.....		40.0		8.5		48.5		

TABLE IXb  
MNSO<sub>4</sub> SET—SECOND CROP—GREENHOUSE SOIL

	MNSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	500	14.05	14.05	3.45	3.45	17.5	17.50	10.5	10.25	28.0	27.75	-4.39
2	500	14.05		3.45		17.5		10.0		27.5		
3	1000	16.50	19.45	3.50	3.50	20.0	22.45	7.5	9.25	27.5	26.70	-5.44
4	1000	22.40		3.50		24.9		11.0		25.9		
5	1500	13.40	12.95	4.40	4.70	17.8	17.65	7.5	6.75	25.3	24.40	-7.74
6	1500	12.50		5.00		17.5		6.0		23.5		
7	2000	13.55	12.94	4.25	3.72	17.8	17.65	10.7	12.00	28.5	29.65	-2.49
8	2000	12.32		5.18		17.5		13.3		30.8		
9	2500	11.77	11.89	3.73	3.87	15.5	15.75	12.5	11.60	28.0	27.35	-4.78
10	2500	12.00		4.00		16.0		10.7		26.7		
11	3000	13.33	13.32	4.67	4.19	18.0	17.50	9.7	9.35	25.7	25.75	-6.38
12	3000	13.30		3.70		17.0		9.0		26.0		
13*	3500	7.50	10.24	3.30	4.41	10.8	14.65	6.5	8.25	17.3	22.90	-9.24
14	3500	12.97		5.53		18.5		10.0		28.5		
15	4000	14.67	15.83	2.33	2.33	17.0	17.00	8.3	9.15	25.3	26.15	-5.99
16	4000	17.00		.....		17.0		10.0		27.0		
17	4500	12.02	16.80	4.98	4.20	17.0	21.00	9.5	7.75	26.5	29.00	-3.14
18	4500	21.58		3.42		25.0		6.0		31.5		
19	5000	13.56	16.53	4.64	4.64	18.2	18.85	9.8	7.40	28.0	26.25	-5.89
20	5000	19.50		.....		19.5		5.0		24.5		
21	5500	18.20	18.20	2.50	2.50	20.7	20.70	9.2	9.20	29.7	29.70	-2.44
22	5500	.....		.....		.....		.....		.....		
23	6000	21.70	24.03	1.30	.92	23.0	24.95	10.0	7.50	33.0	32.45	+0.31
24	6000	26.36		.54		26.9		5.0		31.9		
25	Control	13.08	13.71	5.05	5.16	18.13	18.88	14.8	13.27	32.93	32.14	
26	Control	12.87		4.13		17.0		13.0		30.0		
27	Control	15.17		6.31		21.5		12.0		33.5		

\* One plant died.

TABLE IXc  
MNSO, SET—THIRD CROP—GREENHOUSE SOIL

	MnSO <sub>4</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	500	11.90	11.90	4.70	4.70	16.60	16.60	1.40	1.40	18.00	18.00	+0.57
2*	500	.....		.....		.....		.....		.....		
3	1000	9.54	10.02	3.96	3.98	13.50	14.00	2.00	2.35	15.50	16.35	-1.08
4	1000	10.50		4.00		14.50		2.70		17.20		
5	1500	15.76	13.13	4.24	4.67	20.00	17.80	1.50	1.25	21.50	19.05	+1.62
6	1500	10.50		5.10		15.60		1.00		16.60		
7	2000	21.30	17.20	4.10	3.50	25.40	20.70	1.40	1.50	26.80	22.20	+4.77
8	2000	13.10		2.90		16.00		1.60		17.60		
9	2500	20.15	18.00	5.85	4.50	26.00	22.50	2.60	2.58	28.60	25.08	+7.65
10	2500	15.85		3.15		19.00		2.55		21.55		
11	3000	17.80	16.05	5.00	3.85	22.80	19.90	1.40	1.85	24.20	21.75	+4.33
12	3000	14.30		2.70		17.00		2.30		19.30		
13	3500	13.70	12.95	4.90	4.35	18.60	17.30	1.50	2.05	20.10	19.35	+1.92
14	3500	12.20		3.80		16.00		2.60		18.60		
15	4000	12.25	12.25	4.15	4.15	16.40	16.40	3.50	3.50	19.90	19.90	+2.47
16	4000	.....		.....		.....		.....		.....		
17	4500	12.44	12.40	3.76	3.70	16.20	16.10	2.40	2.40	18.60	18.50	+1.07
18	4500	12.36		3.64		16.00		2.40		18.40		
19	5000	12.40	11.87	3.20	3.83	15.60	15.70	3.20	2.70	18.80	18.40	+0.97
20	5000	11.35		4.45		15.80		2.20		18.00		
21	5500	9.80	9.80	4.40	4.40	14.20	14.20	2.80	2.80	17.00	17.00	-0.43
22	5500	.....	.....	.....		.....		.....		.....		
23	6000	9.64	12.52	4.36	3.48	14.00	16.00	2.10	2.30	16.10	18.30	+0.87
24	6000	15.40		2.60		18.00		2.50		20.60		
25	Control	12.25	12.80	3.25	2.73	15.50	15.53	1.40	1.90	16.90	17.43	
26	Control	13.30		2.30		15.60		2.30		17.90		
27	Control	12.85		2.65		15.50		2.00		17.50		

\* Contaminated by leaky roof.

TABLE Xa  
 MNCL<sub>2</sub> SET—FIRST CROP—GREENHOUSE SOIL

	MnCl <sub>2</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	500	60.0	58.00	.....	.....	60.0	58.00	12.5	11.75	72.5	69.75	+16.62
2	500	56.0		.....		56.0		11.0		67.0		
3	1000	68.2	73.60	.....	.....	68.2	73.60	13.5	12.00	81.7	85.60	+33.47
4	1000	79.0		.....		79.0		10.5		89.5		
5	1500	57.2	65.60	.....	.....	57.2	65.60	13.5	12.50	70.7	78.10	+25.97
6	1500	74.0		.....		74.0		11.5		85.5		
7	2000	37.0	37.55	.....	.....	37.0	37.55	5.9	6.95	42.9	44.50	— 7.63
8	2000	38.1		.....		38.1		8.0		46.1		
9	2500	32.0	35.60	.....	.....	32.0	35.60	12.5	11.75	44.5	47.35	— 4.78
10	2500	39.2		.....		39.2		11.0		50.2		
11	3000	26.0	28.10	.....	.....	26.0	28.10	6.0	6.75	32.0	34.85	—17.28
12	3000	30.2		.....		30.2		7.5		37.7		
13	2500	25.2	22.10	.....	.....	25.2	22.10	4.0	3.25	29.2	25.35	—26.78
14	3500	19.0		.....		19.0		2.5		21.5		
15*	4000	20.0	20.00	.....	.....	20.0	20.00	1.0	1.00	21.0	21.00	—31.13
16*	4000											
17	4500	14.0	14.45	.....	.....	14.0	14.45	1.5	1.30	15.5	15.75	—36.38
18	4500	14.9		.....		14.0		1.1		16.0		
19	5000	12.9	12.90	.....	.....	12.9	12.90	1.2	1.20	14.1	14.10	—38.03
20*	5000	.....		.....								
21	5500	4.5	6.70	.....	.....	4.5	6.70	1.0	1.05	5.5	7.75	—44.38
22	5500	8.9		.....		8.9		1.1		10.0		
23	6000	5.2	3.55	.....	.....	5.2	3.55	.7	.45	5.9	4.00	—48.13
24	6000	1.9		.....		1.9		.2		2.1		
25	Control	41.4	41.46	.....	.....	41.4	41.46	12.5	10.66	53.9	52.13	
26	Control	43.0		.....		43.0		11.0		54.0		
27	Control	40.0		.....		40.0		8.5		48.5		

\* Plants died during growing period.

TABLE Xb  
MnCl<sub>2</sub> SET—SECOND CROP—GREENHOUSE SOIL

	MnCl <sub>2</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	500	14.13	13.07	3.87	3.87	18.00	15.00	8.0	8.25	26.00	23.25	-8.89
2	500	12.00		.....		12.00		8.5		20.50		
3	1000	12.72	10.94	4.98	4.67	17.70	15.60	8.8	8.65	26.50	24.25	-7.89
4	1000	9.15		4.35		13.50		8.5		22.00		
5	1500	15.15	15.80	4.75	4.75	19.90	20.35	8.2	7.70	28.10	28.05	-4.09
6	1500	16.05		4.75		20.80		7.2		28.00		
7	2000	25.08	23.12	4.42	4.73	29.50	27.85	6.9	7.95	36.40	35.80	+3.66
8	2000	21.16		5.04		26.20		9.0		35.20		
9	2500	22.00	20.15	5.00	6.45	27.00	26.60	8.0	8.50	35.00	35.10	+2.96
10	2500	18.30		7.90		26.20		9.0		35.20		
11	3000	26.05	23.55	3.95	4.30	30.00	27.85	10.0	7.60	40.00	35.45	+3.31
12	3000	21.04		4.66		25.70		5.2		30.90		
13	3500	20.66	22.83	1.34	1.92	22.00	24.75	9.0	7.75	31.00	32.50	+0.36
14	3500	25.00		2.50		27.50		6.5		34.00		
15	4000	32.07	33.04	.93	1.46	33.00	34.50	6.4	6.20	39.40	40.70	+8.56
16	4000	34.00		2.00		36.00		6.0		42.00		
17	4500	30.40	27.80	.....	.....	30.40	27.80	6.0	5.70	36.40	33.50	+1.36
18	4500	25.20		.....		25.20		5.4		30.60		
19	5000	29.60	26.05	.90	.90	30.50	26.50	6.5	5.25	37.00	31.75	-0.39
20	5000	22.50		.....		22.50		4.0		26.50		
21	5500	27.03	28.70	.47	.47	27.50	29.00	6.0	5.50	33.50	34.50	+2.36
22	5500	30.50		.....		30.50		5.0		35.50		
23	6000	24.05	27.04	.45	.46	24.50	27.75	6.0	5.50	30.50	33.25	+1.11
24	6000	30.03		.47		31.00		5.0		36.00		
25	Control	13.08	13.71	5.05	5.16	18.13	18.88	14.8	13.20	32.93	32.14	
26	Control	12.87		4.13		17.00		13.0		30.00		
27	Control	15.17		6.31		21.50		12.0		33.50		

TABLE Xc  
MnCl<sub>2</sub> SET—THIRD CROP—GREENHOUSE SOIL

	MnCl <sub>2</sub> added to soil in parts per 1,000,000	Weight of straw gm.	Average weight of straw gm.	Weight of grain gm.	Average weight of grain gm.	Total weight of dry matter above surface gm.	Average weight of dry matter above surface gm.	Weight of roots gm.	Average weight of roots gm.	Total weight of dry matter produced gm.	Average weight of total dry matter produced gm.	Average total difference over control
1	500	12.68	12.97	2.76	4.52	17.20	18.40	1.40	1.63	18.60	20.03	+ 2.60
2	500	13.25		6.35		19.60		1.85		21.45		
3	1000	16.46	15.23	5.74	5.12	22.20	20.35	1.75	1.83	23.95	22.18	+ 4.75
4	1000	14.00		4.50		18.50		1.90		20.40		
5	1500	12.36	12.40	6.06	4.61	18.40	17.00	2.30	2.05	20.70	19.05	+ 1.62
6	1500	12.45		3.15		15.60		1.80		17.40		
7	2000	17.60	17.93	6.00	4.87	23.60	22.80	2.20	2.70	25.80	25.50	+ 8.07
8	2000	18.25		3.75		22.00		3.20		25.20		
9	2500	14.02	13.89	6.28	5.56	20.30	19.45	2.70	2.55	23.00	22.00	+ 4.57
10	2500	13.76		4.84		18.60		2.40		21.00		
11	3000	21.30	16.90	3.70	3.40	25.00	20.30	2.00	2.60	27.00	22.90	+ 5.47
12	3000	12.50		3.10		15.60		3.20		18.80		
13	3500	14.64	18.12	5.16	5.18	19.80	23.30	2.30	2.95	22.10	26.25	+ 8.82
14	3500	21.60		5.20		26.80		3.60		30.40		
15	4000	17.00	18.60	4.00	5.10	21.00	23.70	1.00	1.90	22.00	25.60	+ 8.17
16	4000	20.20		6.20		26.40		2.80		29.20		
17	4500	23.00	20.30	3.80	4.10	26.80	24.40	1.20	1.20	28.00	25.60	+ 8.17
18	4500	17.60		4.40		22.00		1.20		23.20		
19	5000	27.70	24.65	3.90	4.15	31.60	28.80	1.80	1.90	33.40	30.70	+13.27
20	5000	21.60		4.40		26.00		2.00		28.00		
21	5500	17.36	16.03	5.24	2.97	22.60	19.00	.....	.70	22.60	19.35	+ 1.92
22*	5500	14.70		.70		15.40		.70		16.10		
23	6000	26.50	20.60	8.90	6.28	35.40	27.70	.40	.35	35.80	28.05	+10.62
24	6000	16.35		3.65		20.00		.30		20.30		
25	Control	12.25	12.80	3.25	2.73	15.50	15.53	1.40	1.90	16.90	17.43	
26	Control	13.30		2.30		15.60		2.30		17.90		
27	Control	12.85		2.65		15.50		2.00		17.50		

\* Contaminated by leaky roof.





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